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13. SUPPLEMENTARY NOTES

14. ABSTRACT

This report describes year 4 progress of a multi-center study of neurosurgical TBI patients to determine the effects of spreading depression (SD), measured by electrocorticography, on neurologic outcome. In this year, we have enrolled 42 patients, bringing the study total to 136. Data collection, scoring, and monitoring has been completed for 90 patients. Using data from the present study combined with the pilot study, we published the finding that SD is independently associated with worse outcomes in 103 patients. These results establish SD as a mechanism of secondary injury. These analyses will be repeated with the full cohort from the present study and an advanced prognostic model will be developed based on more refined metrics of SD burden, secondary insults, and TBI pathoanatomy. Overall progress indicates that SD should be targeted therapeutically in a future interventional trial. We also found evidence that SD can be monitored non-invasively by scalp EEG recordings, thus providing a method to extend results to non-surgical TBI patients.

15. SUBJECT TERMS
Traumatic brain injury; spreading depression; electroencephalography; intracranial pressure; hypotension;

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I. INTRODUCTION

Severe TBI often results in delayed complications and deterioration of a patient's condition after initial stabilization. This secondary injury is life-threatening and a principal cause of permanent disabilities. *Spreading depressions* (SD) are waves of mass neuronal/astrocytic depolarization that actively propagate a breakdown of ion homeostasis and perturbations of blood flow through injured, but potentially salvageable brain tissue. A large body of pre-clinical research has shown that SD causes expansion of ischemic brain lesions and may be similarly detrimental after TBI. In our preliminary studies using electrocorticography (ECoG) to measure SD in human TBI, we have found that SD occurs in an alarming 58% of patients (18 of 31). *The objective of this study is to determine whether SDs cause secondary injury after TBI, and therefore represent an acute target for treatment to improve recovery.* The Specific Aims of the study are to determine the extent to which spreading depressions are correlated with worse neurologic outcome (6 mo. GOS-E), cerebral metabolism, and secondary physiologic insults (hypotension, high intracranial pressure, and fever).

These aims will be achieved with a prospective, observational clinical study design. Approximately 180 total patients undergoing neurosurgery for treatment of TBI will be enrolled. Subdural electrode strips will be placed during surgery and ECoG recordings will be made for at least 72 hr to measure SD events. Clinical outcome will be assessed at six months according to the extended Glasgow Outcome Scale (GOS-E). Metrics of SD activity will be assessed for their relationship with the GOS-E using a proportional odds model. This regression analysis will include standard co-variates to control for the effects of known factors that independently influence outcome, such as age, GCS score at admission, CT classification, and pupil reactivity. Physiologic variables will be acquired in co-registration with ECoG data. Relationships of SD activity with these measures will be assessed by correlations and comparisons of means and proportions.

The following abbreviations for the study sites are used throughout this report:

University of Pittsburgh	PIT
University of Miami	MIA
Virginia Commonwealth University	VCU
King's College Hospital	KCH
University of Cincinnati	UCCC
Coordinating Center	

II. BODY

A. PATIENT ENROLLMENT and ECoG SCORING

As stated in the Year 3 Annual Report, we aim to enroll an adjusted total of 160 patients by June 30, 2013. Progress in Year 4 has moved us well toward that goal. We enrolled 42 patients which, added to the previous total, yields a total enrollment of 136 patients. Enrollment of 24 additional patients in 10 months is achievable. See Table 1 below.

TABLE 1. Enrollment rates.

	Site	'09	'10	'11	'12	Site Total	Total	Expected 10/12-6/13
Actual	UCCC	6	4	8	17	35		12
	VCU	2	6	3	6	17		4
	MIA	4	6	5	5	20	136	4
	PIT	8	17	12	10	47		8
	KCH	0	8	5	4	17		4
Yearly total		20	41	33	42			

Review of ECoG Recordings

Of the 136 patient recordings reviewed, 17 are unusable due to poor quality/technical failures, or because patients died prior to the start of monitoring. Of the remaining 119 patients, 90 have been formally scored and reviewed.

B. TASK 1: ASSOCIATION OF DEPOLARIZATIONS WITH LONG-TERM OUTCOME

Our analysis of the relationship of spreading depolarizations to 6-month neurologic outcomes in 103 patients has now been published in *Lancet Neurology*. The paper was accompanied by a commentary by Nino Stocchetti. In a subsequent issue, a letter to the editor was published along with our response from authors. These papers are contained in the Appendix of this report.

C. TASK 2: ASSOCIATION OF DEPOLARIZATIONS WITH LOCAL CEREBRAL METABOLISM

Local cerebral blood flow (CBF) is measured by the thermal diffusion technique by placing Hemedex probes adjacent to ECoG electrode strips during surgery. In this reporting period, 5 additional study patients have been monitored with Hemedex, bringing the study total to 21 patients. Of these, 4 patients have now shown CBF transients in association with spreading depolarizations, and 3 of these have been a spreading ischemic reponse, as described in the Year 3 Annual Report. These results are now being analyzed and prepared for publication.

D. TASK 3: SYSTEMIC RISK FACTORS THAT INFLUENCE DEPOLARIZATIONS

Our previous analysis of effects of analgesics and sedation on spreading depolarizations have now been published in *Brain*. The manuscript is contained in the Appendix.

We continue to record systemic physiologic data during ECoG monitoring for assessment of secondary insults and their relationship to spreading depolarizations. Programs have been

written for statistical analysis, which will proceed after enrollment of the final patient and completion of the database.

E. NEW RESULTS

Non-invasive monitoring
A major limitation of the present study and research is that spreading depolarizations can only be monitored using invasive techniques. Thus, the study and application is restricted to patients with severe TBI who undergo surgery. We are therefore investigating whether spreading depolarizations can also be monitored by non-invasive scalp EEG recordings – a technique that can be used with any patient and TBI severity. In association with COSBID, we found that scalp EEG does exhibit clear signatures of depolarizations in patients with sub-arachnoid hemorrhage (Drenckhahn et al., *Brain*, 2011). We have observed the same correlations in TBI patients, as reported in our National Neurotrauma Society abstract (see Appendices).

Spreading depolarizations as a real-time biomarker
In a substudy of two centers (KCH and VCU), we found significant differences in surgical techniques (decompressive craniectomy) used and in patient outcomes. Patients at VCU had significantly earlier and larger craniotomies and better outcomes, despite being similar in initial injury characteristics compared to KCH patients. VCU patients also had a significantly lower incidence of spreading depolarizations. These data were reported at the National Neurotrauma Society meeting (See Appendix) and 1) affirm the association between depolarizations and outcome, and 2) suggest that depolarization monitoring can be used as a biomarker of adverse patient course in evaluation of therapeutic and surgical interventions.

SUMMARY OF MILESTONE AND TASK COMPLETION
The following Table summarizes study progress according to the SOW tasks/milestones. Milestone completion/progress is denoted by green bars.

TIMETABLE FOR THE RESEARCH PROGRAM		Prior	YR1	YR2	YR3	YR4
Task #1:	Spreading depressions as independent predictors of worse neurologic outcome					
Milestone #1.	*Complete regulatory review and obtain approval for human use protocols*	green				
Milestone #2.	*Implementation of online database*		green			
Milestone #3.	*Final protocol briefing by P.I. to each medical center team*		green			
Milestone #4.	*Patient enrollment and electrocorticography*		green	green	green	
Milestone #5.	*Score ECoG recordings and quantify SD activity by defined metrics*			green	green	
Milestone #6.	*Assessment of neurologic outcomes at 6 months post-TBI*			green	green	
Milestone #7.	*Statistical analysis and publication*					green
Task #2:	Association of CBF and spreading depression activity					

Milestone #1.	**Patient enrollment: simultaneous ECoG and CBF measures**					
Milestone #2.	**Data analysis and publication**					
Task #3:	Association of spreading depression and secondary physiologic insults					
Milestone #1.	**Patient enrollment: simultaneous ECoG and physiologic parameter monitoring**					
Milestone #2.	**Write custom data processing programs for analysis**					
Milestone #3.	**Data analysis and publication**					

III. KEY RESEARCH ACCOMPLISHMENTS

● We have demonstrated that *SD is a significant predictor of worse outcome, independent of known predictors for poor outcome at hospital admission* (Hartings et al., *Lancet Neurology* 10(12):1058-1064, 2011).

● We developed a signal processing method to recover full-band DC waveforms from the raw, frequency-limited clinical ECoG recordings. We applied this method to directly assess the duration of cortical tissue depolarization associated with individual SD events and found that a *significant proportion of SD events have pathologically prolonged durations, suggesting that they cause tissue damage. Patients with prolonged SDs had worse outcomes* (Hartings et al., *Brain* 134:1529-40, 2011). Furthermore, we have developed a new recording platform for acquisition of full-band DC recordings, enabling direct assessment of depolarization duration (Wilson et al., *Acta Neurochirugica Supplementum* 115: 67-74, 2013).

● We found a trend toward *increased risk of post-traumatic epilepsy in patients with SD* (unpublished).

● We have found that *SD is significantly associated with excitotoxicity and metabolic crisis, as measured by cerebral microdialysis*. These results suggest a vicious cycle in which metabolic crisis and elevated glutamate trigger SD, which in turn increases metabolic demand and failure with further excitotoxic damage (Wilson et al., *Journal of Neurotrauma* 27(5): A-4, 2010).

● We have found that *SD in TBI can be coupled to pathologic hemodynamic responses in the form of 'cortical spreading ischemia.'* This introduces a new pathophysiologic concept and mechanism in TBI (unpublished).

● We found that *ketamine significantly inhibits SD occurrence*, while other commonly used sedatives and analgesics do not (Hertle et al., *Brain* 135(Pt 8): 2390-2398, 2012). Thus, ketamine is identified as a potential therapeutic agent for TBI.

● We found that the *risk of developing SD is significantly associated with the degree of traumatic subarachnoid hemorrhage*, but is not associated with contusions, intraparenchymal or subdural hemorrhage, midline shift, or status of basal cisterns (Losiniecki et al., *Journal of Neurotrauma* 27(5): A-87, 2010). These results show that TBI pathophysiology varies according to features easily identified by CT, which could be used to select patients for targeted clinical trials.

● We found that *high temperature, low blood pressure, and low cerebral perfusion pressure are associated with increased SD activity*. Dose-response relationships were described (Hartings et al., *Journal of Neurotrauma.* 26(11):1857-1866, 2009). Thus, carefully targeted physiologic values may serve as a therapeutic technique to inhibit SD and improve outcomes.

● We found that a significant portion of SDs can be detected as amplitude depressions in continuous scalp EEG recordings. This finding suggests that *SD can be monitored non-invasively* and provides a method to extend results from the present study to all TBI patients.

IV. REPORTABLE OUTCOMES

Published Articles:

Hartings JA, Bullock MR, Okonkwo DO, Murray L, Murray GD, Fabricius M, Maas AIR, Woitzik J, Sakowitz O, Mathern B, Roozenbeek B, Lingsma H, Dreier JP, Puccio A, Shutter LA, Pahl C, Strong AJ (2011) **Spreading depolarizations and outcome after traumatic brain injury: a prospective, observational study**. *Lancet Neurology* 10(12):1058-1064.

Hertle DN, Dreier JP, Woitzik J, Hartings JA, Bullock MR, Okonkwo DO, Shutter LA, Vidgeon S, Strong AJ, Kowoll C, Dohmen C, Diedler J, Veltkamp R, Bruckner T, Unterberg AW, Sakowitz OW (2012) **Effect of analgesics and sedatives on the occurrence of spreading depolarizations accompanying acute brain injury**. *Brain* 135(Pt 8): 2390-2398.

Hartings JA, Strong AJ, Okonkwo DO, Bullock MR (2012) **Spreading depolarisations and traumatic brain injury: time course and mechanisms – Author's reply**. *Lancet Neurology* 11: 389–90.

Drenckhahn C, Winkler M, Major S, Sheel M, Kang EJ, Grozea C, Hartings JA, Woitzik J, Dreier JP (2012) **Correlates of spreading depolarization in human scalp electroencephalography**. *Brain* 135:853-868.

Wilson JA, Shutter LA, Hartings JA (2013) **A platform for multimodal data collection and research in neurocritical care**. *Acta Neurochirugica Supplementum* 115: 67-74.

Manuscripts in Preparation:

Steve Vidgeon, M. Ross Bullock, Chris Zacko, Thomas Ridder, Rick Stanger, Martin Fabricius, Bruce Mathern, Anthony J. Strong, Jed A. Hartings. **Efficacy of pre-emptive decompressive craniectomy: a comparison between a UK and US neurosurgical centre.** Prepared for *J. Neurosurg.*

Abstracts and Conference Presentations:

Hartings JA, Okonkwo DO, Bullock MR, Shutter LA, Mathern B, Strong AJ. Spreading depolarizations and outcome after traumatic brain injury: a prospective observational study. Military Health System Research Symposium (8 Aug 2012; Ft. Lauderdale, FL).

Hartings JA, Bullock MR, Zacko C, Vidgeon S, Ridder T, Stanger R, Fabricius M, Mathern B, Tolias C, Pahl C, Strong AJ. Pre-emptive decompressive craniectomy: a comparative effectiveness study between a U.K. and U.S. center. National Neurotrauma Society 2012 (Phoenix, AZ).

Hartings JA, Wilson JA, Pollandt S, Andaluz N, Ficker D, Shutter LA. A matter of perspective: detection of spreading depolarizations by continuous EEG. Princeton Conference XXVIII (2012; Cincinnati, OH) and National Neurotrauma Society 2012 (Phoenix, AZ).

Dreier JP, Hartings JA. Spreading convulsions, spreading depolarization, and epileptogenesis in human cerebral cortex. Winter Conference on Brain Research (Jan 2012; Snowbird, Utah)

Seminars at National Meetings:

International Razavi Neurotrauma Congress (Nov 23-25, 2011, Mashhad, Iran): "Spreading depolarization and outcome after traumatic brain injury: prospective, observational study"

Funding Applications Based on Work Supported by This Award:

Title: Spreading depolarizations in acute brain injury
Principal Investigator: Hartings
Funding agency: Established Investigator Award, American Heart Association
Status: Under review

Title: Epileptiform activity leads to increased intracranial pressure in traumatic brain injury
Principal Investigator: P. Vespa (UCLA); Co-Investigator: Hartings
Funding agency: NIH RO1
Status: Not funded

V. CONCLUSION

Here we have reported continuing progress in a challenging emergency medicine study involving the use of invasive monitoring of patients with moderate-to-severe TBI. In this year, we have enrolled an additional 42 patients, bringing the study total to 136. Importantly, a large volume of clinical data is being collected from these patients, including patient outcome, in our electronic case report form (eCRF) database. ECoG data quality is excellent, and patients thus far show a ~50% incidence of SD.

Most critically, we have published results showing an independent association of SD with poor outcome, which represents a major step in completing the overall goal of the study. This was accomplished using a significant number of patients from the pilot study. As outlined in the commentary by Nino Stocchetti (see Appendix 2), the further increase of our sample size through completion of present enrollment will be important to validate our results in a second, non-overlapping cohort and to address limitations of the initial study. These include 1) testing of additional metrics of SD activity to clarify their relationships with outcome and determine the best prognostic ECoG measures, 2) determine the relationship of SD to primary TBI injury types, 3) determine the relationship of SD to secondary insults.

Future course
We aim to enroll 160 total patients by June 30, 2013, as described above. Enrollment will continue to this date, even if 160 patients is exceeded, in order to maximize statistical power. Allowing for 6 months to record patient outcomes, and a final 6 months for study close-out and final analysis and publication of results, the study would therefore terminate on June 30, 2014. Analysis of the budget shows that the study can be extended for this period without additional funds, and therefore a second one-year extension-without-funds will be sought to allow completion of the study through this date.

VI. REFERENCES

Hartings JA, Strong AJ, Fabricius M, Manning A, Bhatia R, Dreier JP, Mazzeo AT, Tortella FC, Bullock MR (2009) Spreading depolarizations and late secondary insults after traumatic brain injury. *Journal of Neurotrauma.* 26(11):1857-1866.

Hartings JA, Watanabe T, Bullock MR, Okonkwo DO, Fabricius M, Woitzik J, Dreier JP, Puccio A, Shutter LA, Pahl C, Strong AJ (2011) Spreading depolarizations have prolonged direct current shifts and are associated with poor outcome in traumatic brain injury. *Brain* 134:1529-40.

Losiniecki AJ, Okonkwo DO, Shutter LA, Bullock MR, Strong AJ, Mathern B, Puccio A, Vidgeon S, Pahl C, Hartings JA (2010) Computed tomographic pathologies associated with cortical dysfunction after traumatic brain injury requiring surgery. Journal of Neurotrauma 27(5): A-87.

Wilson JA, Bullock MR, Hartings JA (2010) Association of spreading depolarizations with excitotoxicity and metabolic crisis in TBI: a clinical microdialysis study. Journal of Neurotrauma 27(5): A-4.

VII. APPENDICES

Appendix 1   Hartings et al., *Lancet Neurology* publication

Appendix 2   Stocchetti *Lancet Neurology* commentary

Appendix 3   Hartings et al., *Lancet Neurology*, letter-to-editor and author reply

Appendix 4   Hertle et al., *Brain* publication

Appendix 5   Drenckhahn et al., *Brain* publication

Appendix 6   Wilson et al., *Acta Neurochir Suppl* publication

Appendix 7   National Neurotrauma Society abstract

Appendix 8   National Neurotrauma Society abstract

Spreading depolarisations and outcome after traumatic brain injury: a prospective observational study

Jed A Hartings, M Ross Bullock, David O Okonkwo, Lilian S Murray, Gordon D Murray, Martin Fabricius, Andrew I R Maas, Johannes Woitzik, Oliver Sakowitz, Bruce Mathern, Bob Roozenbeek, Hester Lingsma, Jens P Dreier, Ava M Puccio, Lori A Shutter, Clemens Pahl, Anthony J Strong, for the Co-Operative Study on Brain Injury Depolarisations

Summary

Background Pathological waves of spreading mass neuronal depolarisation arise repeatedly in injured, but potentially salvageable, grey matter in 50–60% of patients after traumatic brain injury (TBI). We aimed to ascertain whether spreading depolarisations are independently associated with unfavourable neurological outcome.

Methods We did a prospective, observational, multicentre study at seven neurological centres. We enrolled 109 adults who needed neurosurgery for acute TBI. Spreading depolarisations were monitored by electrocorticography during intensive care and were classified as cortical spreading depression (CSD) if they took place in spontaneously active cortex or as isoelectric spreading depolarisation (ISD) if they took place in isoelectric cortex. Investigators who treated patients and assessed outcome were masked to electrocorticographic results. Scores on the extended Glasgow outcome scale at 6 months were fitted to a multivariate model by ordinal regression. Prognostic score (based on variables at admission, as validated by the IMPACT studies) and spreading depolarisation category (none, CSD only, or at least one ISD) were assessed as outcome predictors.

Findings Six individuals were excluded because of poor-quality electrocorticography. A total of 1328 spreading depolarisations arose in 58 (56%) patients. In 38 participants, all spreading depolarisations were classified as CSD; 20 patients had at least one ISD. By multivariate analysis, both prognostic score (p=0·0009) and spreading depolarisation category (p=0·0008) were significant predictors of neurological outcome. CSD and ISD were associated with an increased risk of unfavourable outcome (common odds ratios 1·56 [95% CI 0·72–3·37] and 7·58 [2·64–21·8], respectively). Addition of depolarisation category to the regression model increased the proportion of variance in outcome that could be attributed to predictors from 9% to 22%, compared with the prognostic score alone.

Interpretation Spreading depolarisations were associated with unfavourable outcome, after controlling for conventional prognostic variables. The possibility that spreading depolarisations have adverse effects on the traumatically injured brain, and therefore might be a target in the treatment of TBI, deserves further research.

Funding US Army CDMRP PH/TBI research programme.

Introduction

Traumatic brain injury (TBI) poses a major public-health problem because it is the leading cause of mortality and disability in young people in high-income countries.[1] Fundamental challenges in advancing the treatment of TBI are its heterogeneity in terms of cause, pathology, and severity, and the absence of mechanistic therapeutic targets. The findings of the International Mission on Prognosis and Clinical Trials (IMPACT) studies[2] have provided approaches for dealing with the prognostic heterogeneity in TBI,[3–5] and fairly broad inclusion criteria, covariate adjustment, and ordinal analysis have the potential to increase statistical power in clinical trials by up to 50%.[6] Until now, however, mechanistic targets have been scarce because of the difficulty in monitoring and validation of relevant pathological processes in patients with TBI. Nonetheless, monitoring of mechanisms of tissue damage could help to explain the heterogeneity of TBI, guide treatment decisions, and enable the detection of relevant treatment effects.

Spreading depolarisations are pathological events that recur acutely in the cerebral grey matter of patients with TBI and other acute brain injuries.[7–9] Detected and measured by electrocorticography in the intensive-care unit, they represent a pathological mechanism that might be targeted for monitoring and treatment.[10,11] Spreading depolarisations are a class of slowly propagating cortical waves (2–5 mm/min) that were first described by Leão[12] in 1944 as "cortical spreading depression" and are characterised by near-complete sustained depolarisation of neurons and astrocytes.[11,13] During depolarisation, breakdown of ion gradients, neuronal swelling and dendritic beading, loss of electrical activity, and increased metabolic demand take place; these changes must be matched by adequate substrate delivery to restore membrane potentials and prevent neuronal injury by mitochondrial damage, intracellular calcium (Ca^{2+}) accumulation, and excitotoxicity.[10] Since the 1990s, spontaneous waves of peri-infarct spreading depolarisations have been appreciated as a central mechanism of the penumbral expansion of cerebral infarcts.[14] Waves

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can be initiated when the extracellular concentration of potassium (K^+) exceeds a threshold of roughly 12 mmol/L, as in the ischaemic core, or when hyperexcitable foci develop in other disease models.

Findings of clinical studies have translated this large body of published experimental work, showing that spreading depolarisations happen spontaneously and abundantly in malignant stroke, subarachnoid haemorrhage, and TBI in human beings. Evidence suggests that spreading depolarisations have an adverse effect on injured human cerebral cortex,[15–18] are affected by systemic perfusion, glucose, temperature, and drugs,[9,19] and might provide a mechanistic therapeutic target and real-time biomarker to guide treatment. To investigate this potential, we must ascertain whether the occurrence of spreading depolarisations is associated with a clinically meaningful endpoint and provides information not available in other assessments.

We have previously reported, by univariate analysis, that spreading depolarisations are associated with unfavourable 6-month outcomes after TBI.[20] However, spreading depolarisations might be only a marker of injury severity, and confounding covariates could account for some or all of the noted association. An independent relation of spreading depolarisations with outcome, on the other hand, would provide evidence that depolarisations are a causal factor affecting outcome. Our aim with the Co-Operative Study on Brain Injury Depolarisations was to test the null hypothesis that spreading depolarisations have no independent association with 6-month outcomes, using multivariate analysis to control for established prognostic factors.

Methods

Patients

We undertook a multicentre study at seven neurosurgical centres. We prospectively enrolled patients with acute TBI who met the following inclusion criteria: clinical decision for craniotomy for lesion evacuation, decompression, or both; surgery fewer than 7 days after trauma; and age 18 years or older. We excluded individuals with fixed dilated pupils. The first group of patients formed a pilot phase and a subsequent group was enrolled in a second phase, funded by the US Army, which is registered at ClinicalTrials.gov, number NCT00803036. Participants included patients analysed in two previous studies.[9,20]

The institutional review boards of every site approved the research, which was undertaken in accordance with the Declaration of Helsinki. Owing to the nature of brain injury, patients were unable to provide study consent and, therefore, written informed consent was obtained from legally authorised representatives before the start of all study procedures.

Procedures

We gathered data prospectively for seven variables on the basis of patients' characteristics at admission, as covariates

for outcome prediction (defined by the IMPACT study).[5,21] Variables were age, Glasgow coma scale motor score, pupillary reactivity, Marshall CT category, and the presence of hypotension, hypoxia, and traumatic subarachnoid haemorrhage. We used the motor and pupillary responses that were most representative of the post-resuscitation condition. We ascertained hypotension and hypoxia for the period before or at admission (ie, at the accident scene, during transport to hospital, or at the first treating hospital) from emergency medical service records or information from previous hospitals. We judged patients to be hypotensive if systolic blood pressure was less than 90 mm Hg and hypoxic if their partial arterial pressure of oxygen was less than 60 mm Hg, if their saturation level of oxygen in blood was less than 90%, or on the basis of strong clinical suspicion. We based CT scoring on scans at admission. In the initial pilot phase of this study, we did not obtain detailed data prospectively on indications for surgery and surgical procedures undertaken. However, this information was gathered for some individuals from the pilot phase and all enrolled in the second study phase (webappendix p 1).

At the end of surgery, we placed an electrode strip on the surface of the cortex for subsequent electrocorticography.[7,8] We positioned the strip near the injury focus on viable but often oedematous or contused cortex with a low load of subarachnoid blood. We transferred patients to the intensive-care unit after surgery, where we initiated continuous electrocorticography. Neurocritical care practices accorded with guidelines for management of severe TBI.[22] We did not specify any further neurocritical care protocols. Preferred sedatives and analgesics were fentanyl and its analogues, benzodiazepines, and morphine. We discouraged use of propofol or thiopental, although analysis of clinical data suggests they do not affect spreading depolarisations (Daniel Hertle, Heidelberg University Hospital, Heidelberg, Germany; personal communication). Clinicians were masked to electrocorticographic results and no treatment decisions were based on recordings. When invasive monitoring was no longer needed clinically, or after a maximum of 7 days, we ended electrocorticography and removed electrode strips at the bedside by gentle traction. We assessed neurological outcome at 6 months according to the extended Glasgow outcome scale (eGOS) by a telephone interview or clinic visit. Assessors were unaware of electrocorticographic results.

Electrocorticographic recordings were made from the linear subdural strip, which consisted of six electrodes with 10 mm spacing between contacts (Wyler, Ad-Tech Medical, Racine, WI, USA). We connected electrodes in a sequential bipolar fashion to AC-coupled amplifiers (Dual Bioamp or Octal amplifiers, ADInstruments, NSW, Australia; GT205, Guger Technologies, Graz, Austria) with 0·02 Hz or 0·01 Hz high-pass cutoff. We recorded data and reviewed them with 200 Hz sampling by a Powerlab 16/SP and LabChart software, version 7.2 (ADInstruments).

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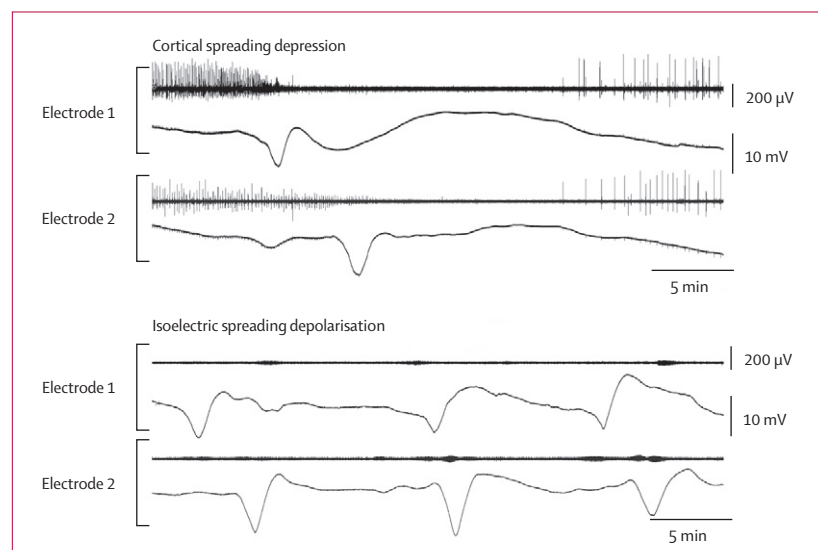
Figure: Spreading depolarisations with depression of spontaneous activity and in isoelectric cortex
Recordings from subdural electrodes show high-frequency components in upper traces (high-pass, filtered at 0·5 Hz) and low-frequency components in lower traces (full-band). Electrodes 1 and 2 are 10 mm apart. Both types of depolarisation show negative shifts of the slow potential that propagate from electrode 1 to electrode 2 at 1–5 mm/min. In the case of cortical spreading depression, onset of slow-potential change coincides with depression of spontaneous cortical activity, which begins recovery after 15 min. In the case of isoelectric spreading depolarisation, no spontaneous cortical activity takes place.

We analysed recordings offline for spreading depolarisations, according to methods established during interim analysis of initial study data.[8] Briefly, the signature of spreading depolarisation is a slow-potential change of 1–5 mV peak-to-peak amplitude in the near-DC (<0·1 Hz) frequency band, reflecting in part the intracellular flux of cations during mass tissue depolarisation that propagates at 1–6 mm/min between electrodes. When spontaneous brain electrical activity is present (0·5–100 Hz), the depolarisation also causes electrocorticographic depression, which spreads with the slow-potential. Thus, we identified depolarisations by spreading slow-potential changes and classified them either as isoelectric spreading depolarisation (ISD), when no spontaneous activity was present, or as cortical spreading depression (CSD), when depolarisation caused depression of spontaneous electrocorticographic activity (figure).

The distinction between ISD and CSD has been made in previous clinical studies of TBI (n=53 patients),[20] ischaemic stroke (n=16),[23] and aneurysmal subarachnoid haemorrhage and spontaeous intracerebral haemorrhage (n=12).[8] This distinction is a dichotomisation of the continuum of depolarisation characteristics as observed in clinical and experimental studies. In focal ischaemia, for instance, spreading depolarisations arise at the rim of the infarct core and either propagate perpendicular to gradients of increasing perfusion, oxygen, and glucose through the penumbra into healthy tissue or remain in the penumbra and cycle around the core.[14,17] When traversing the penumbra, these peri-infarct depolarisations are prolonged and cause lesion growth as further tissue is terminally depolarised,[24,25] whereas necrosis is not observed in remote tissue, where shorter depolarisations cause transient depression of spontaneous activity. Because the ischaemic penumbra is defined as functionally silent (isoelectric) but viable (ie, no persistent steep elevation of extracellular K^+),[26,27] ISD in traumatically injured brain might be analogous to peri-infarct depolarisation, with similar pathogenic effects. Analysis of 295 events in patients with TBI confirmed that ISD causes more prolonged tissue depolarisation than does CSD.[20] Thus, we categorised patients' recordings as one of three types: ISD if at least one ISD arose, with or without additional CSD; CSD if only CSD and no ISD happened; or no depolarisations.

Statistical analysis

Pilot data available at study inception were inadequate for statistical powering.[7] Rather, we decided to analyse data addressing the central hypothesis with a proportional odds model after enrolment of 100 patients, a lower limit based on practical difficulties of enrolling such a select group of patients. This sample size was too small to allow a full multivariate prognostic model to be fitted. Therefore, we used the linear predictor from the published seven-variable model to capture the effect of the conventional covariates in a summary prognostic score.[21] For instance, prognostic scores of –2·2, 0·0, and 2·2 correspond to 10%, 50%, and 90% probabilities of unfavourable outcome (eGOS 1–4). In effect, we used the large sample of patients analysed by Hukkelhoven and colleagues[21] to give an appropriate weighting of these seven conventional variables. A few covariate values (16 [1·9%] of 824) were missing and were imputed using their expected values conditional on the recorded covariates and the eGOS for the relevant patients.

For primary analysis of the eGOS, we used ordinal regression, assuming a proportional odds model. We fitted the model with depolarisations as a categorical variable and with prognostic score as a continuous linear term. We based model comparisons on a likelihood ratio

	No depolarisation (n=45)	Cortical spreading depression (n=38)	Isoelectric spreading depolarisation (n=20)
Dead*	9 (20%)	7 (18%)	12 (60%)
Vegetative*	1 (2%)	5 (13%)	0
Lower severe disability*	4 (9%)	5 (13%)	3 (15%)
Upper severe disability*	5 (11%)	3 (8%)	2 (10%)
Lower moderate disability †	8 (18%)	4 (11%)	2 (10%)
Upper moderate disability †	7 (16%)	4 (11%)	0
Lower good recovery †	5 (11%)	6 (16%)	1 (5%)
Upper good recovery†	6 (13%)	4 (11%)	0

Percentages indicate the proportion of patients relative to the total number in the respective depolarisation category. Percentage totals do not equal 100% due to rounding. eGOS=extended Glasgow outcome score. *Outcomes were classified as unfavourable (eGOS 1–4). †Favourable outcomes (eGOS 5–8).

Table 1: Distribution of eGOS at 6 months by depolarisation category

test. We used ordinal regression because dichotomisation of the eGOS scale discards relevant information and is statistically inefficient.[28] We used Kruskal-Wallis and χ^2 tests for univariate analyses, and we judged $p<0.05$ to be significant.

Role of the funding source

The funding source had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all data in the study and had final responsibility for the decision to submit for publication.

Results

Between October, 2004, and February, 2010, a total of 109 patients were enrolled, 75 into the pilot phase (October, 2004, to February, 2009) and 34 into the second study phase (February, 2009, to February, 2010); six individuals were excluded from the pilot study owing to poor quality of electrocorticography. 76 (74%) patients were men; 97 (94%) had closed head injuries from falls, motor vehicle and cycling accidents, and assaults, and the remainder had penetrating injuries. All patients underwent craniotomies for evacuation of intracranial mass lesions or cerebral decompression, a median of 9·9 h (IQR 4·5–26·3) after trauma. Electrocorticography commenced 4·5 h (3·2–7·8) later, after transfer to intensive care, and continued for 67 h (40–102).

In the 74 patients for whom data were collected, 41 (55%) surgical procedures were indicated by initial CT findings, whereas the remaining 33 (45%) surgical procedures were undertaken after development of subsequent complications (webappendix p 1). Predominant lesion types were acute subdural haematomas and contusions. Lateralised decompressive craniectomies were frequently done together with lesion evacuation, which in 28 (38%) individuals entailed re-section of contused brain. No haemorrhagic or infectious complications were associated with the electrode strip.

A total of 1328 spreading depolarisations were recorded in 58 (56%) of 103 patients. Of these, 20 (19%) patients had at least one ISD, with or without additional CSD, and 38 (37%) had CSD only. The remaining 45 patients (44%) had no spreading depolarisations. Table 1 shows the distribution of 6-month neurological outcomes (eGOS) in these three subgroups. 17 (85%) patients with ISD, 20 (53%) with CSD, and 19 (42%) with no spreading depolarisations had unfavourable outcomes (p=0·006). Neither recording duration nor the timing of surgery post-trauma differed between groups (p=0·82 for both). Seizures were also recorded with electrocorticography in 14 patients, including 11 (19%) of the 58 with depolarisations and three (7%) of the 45 without (p=0·07).

Table 2 shows the seven conventional covariates for outcome prediction that were used to calculate patients' prognostic scores. Median prognostic scores were 0·06 (IQR −0·20 to 0·98) for patients who had ISD,

	Participants (n=103)
Age (years)	44 (30 to 60, 18 to 79)
Glasgow coma scale motor score*	
No response	24 (23%)
Extension	4 (4%)
Flexion	5 (5%)
Withdraws	18 (17%)
Localises	30 (29%)
Obeys	22 (21%)
Pupils*	
Both reacting	66 (64%)
One reacting	16 (16%)
Neither reacting	21 (20%)
Marshall CT category*	
Diffuse injury (2)	3 (3%)
Swelling (3)	4 (4%)
Shift (4)	3 (3%)
Evacuated mass lesion (5)	63 (62%)
Non-evacuated mass lesion (6)	29 (28%)
Traumatic subarachnoid haemorrhage	83 (81%)
Hypotension*	15 (15%)
Hypoxia*	19 (18%)
Prognostic score	0·1 (−0·5 to 1·2, −2·0 to 3·1)

Data are n (%) or median (IQR, range). *16 (1·9%) of the 824 covariate values were missing and were replaced by imputation, comprising four values for Glasgow coma scale motor score, one for pupillary reactivity, six for hypotension, four for hypoxia, and one for Marshall CT category.

Table 2: Covariates for outcome prediction

−0·15 (−0·68 to 0·90) for those who had CSD only, and 0·57 (−0·42 to 1·28) for those with no depolarisations, which did not differ significantly (p=0·34). Similarly, no individual covariate was associated significantly with depolarisation category. For instance, five (50%) of ten patients with diffuse injury, swelling, or shift (Marshall CT categories 2–4) had spreading depolarisations. Patients with mass lesions that were surgically evacuated or not evacuated had similar incidences of spreading depolarisation, with 35 (56%) of 63 and 18 (62%) of 29, respectively (p=0·76).

Prognostic scores were used in multivariate analysis to assess the independent predictive value of spreading depolarisations. Depolarisation category added value for outcome prediction over and above the prognostic score based on conventional outcome predictors (table 3). In particular, the occurrence of ISD was associated with an increased risk of unfavourable outcome (common odds ratio 7·58, 95% CI 2·64–21·8; p=0·0002), whereas no significant association was seen for CSD (1·56, 0·72–3·37; p=0·26). Addition of depolarisation category as a covariate to a model that included only the prognostic score increased the explained variance in outcome (Nagelkerke R^2) from 9% to 22%. The occurrence of spreading depolarisations, irrespective of ISD or CSD category, was also independently associated with increased

	Common odds ratio (95% CI)	p
Prognostic score	1·76 (1·26–2·46)	0·0009
Depolarisation		0·0008
None	1·0	..
CSD	1·56 (0·72–3·37)	0·26
ISD	7·58 (2·64–21·8)	0·0002
CSD=cortical spreading depression. ISD=isoelectric spreading depolarisation.		

Table 3: Outcome prediction (multivariate ordinal regression analysis)

risk of an unfavourable outcome (common odds ratio 2·55, 95% CI 1·25–5·20; p=0·01). Thus, the prognostic value of spreading depolarisations is not dependent on the occurrence of isoelectricity or the particular method used to categorise spreading depolarisations.

Repeating this analysis using only the 91 patients with complete covariate data yielded similar results. Adjusted common odds ratios were 1·67 (95% CI 0·74–3·78; p=0·22) for CSD and 9·01 (2·94–27·6; p=0·0001) for ISD. Analysis of the full cohort of 103 patients using binary logistic regression for the dichotomised eGOS also gave similar results: adjusted odds ratios were 1·85 (0·74–4·65; p=0·189) for CSD and 8·58 (2·13–34·6; p=0·003) for ISD. For further sensitivity analysis, other multivariate models were tested using only the strongest predictors among the covariates; parameter estimates for depolarisations changed little and significance was not affected.

Presence of seizures was not significantly associated with more unfavourable outcomes when added to the multivariate model in table 3 (common odds ratio 2·33, 95% CI 0·75–7·22; p=0·14). Addition of enrolment centre to the model did not affect the significance of other variables, and the centre variable itself was not associated with outcome (p=0·69).

Discussion

Our findings show that spreading depolarisations are associated robustly with unfavourable eGOS scores at 6 months, independent of baseline outcome predictors including age, Glasgow coma scale, and pupillary reactivity. Furthermore, depolarisation categories accounted for 13% of outcome variance beyond that accounted for by established prognostic factors. Surprisingly, prognostic scores for patients with and without spreading depolarisations did not differ, but showed some evidence of worse prognoses for patients without spreading depolarisations. Together, our results provide evidence that spreading depolarisations have an adverse independent effect on clinical outcome and provide information not available in routine prognostic assessments. Spreading depolarisations, therefore, are a candidate mechanism for monitoring and a potential therapeutic target.

Intracerebral monitoring has been a cornerstone of management of severe TBI for decades. The focus has been on maintenance of adequate cerebral blood flow and oxygenation, and intracranial pressure is monitored for calculation of cerebral perfusion pressure and as an indicator of mass lesions and worsening intracranial pathology. Brain tissue oximetry and microdialysis provide additional focal measures of metabolic status and cerebral neurochemistry. Our findings add to this repertoire by showing the clinical relevance of spreading depolarisations, which have been characterised extensively in animal models and can now be monitored in clinical populations. Electrocorticographic monitoring of spreading depolarisations captures distinct cortical events associated with collapse of ionic homoeostasis, intracellular Ca^{2+} influx, cellular oedema, and increased metabolism. The independent association of spreading depolarisations with unfavourable outcomes, after controlling for prognostic variables and injury severity, provides strong evidence that these events could have an adverse effect in human TBI. However, we emphasise that only a positive result from an interventional trial targeting spreading depolarisations could provide definitive proof of a causal effect of spreading depolarisations on outcome.

At present, stratification of patients with TBI with respect to treatments, clinical trials, and outcome prediction is based mainly on factors that can be recorded on admission to specialist care. The predictive value of these factors—principally admission demographics, neurological examination, and CT scan—has been analysed extensively in a series of papers from the IMPACT group[2,4,5] and served as a control variable (prognostic score) in our analysis. Other factors affecting outcome are secondary insults that might arise after admission to specialist care, including hypotension, reduced brain oxygenation, raised intracranial pressure, and pyrexia.[29] These factors do not have a standard measure for quantification, but should also be thought of as potential confounding variables that could affect spreading depolarisation incidence. For example, we noted that temperature and arterial pressure affected the rate of spreading depolarisations over a broad range, although only 8% of all spreading depolarisations were associated with a mean arterial pressure below 70 mm Hg.[9] Thus, spreading depolarisations might not be truly independent, but rather be associated with raised intracranial pressure or low perfusion pressures. Standard methods for measuring the burden of secondary insults are being developed for inclusion in future prognostic modelling. Nonetheless, our results suggest that spreading depolarisations could be, at the very least, important mediators of the effects of secondary insults and hence an attractive target for treatment (panel). Other intensive-care variables, such as sedatives and plasma glucose control, are also under study (eg, JAH is investigating plasma glucose) and are vital to the understanding of why spreading depolarisations occur and how their monitoring might inform management of TBI.

We used a simple method to distinguish two types of spreading depolarisation. However, spreading depolarisations are a class of events along a pathogenic continuum,

with effects that can vary from complete recovery to pan-necrotic cortical lesions. At one end, short-lasting depolarisations (about 1–3 min) elicited in intact cortex induce depression periods of less than 10 min and provoke transient hyperaemia in the cortical microcirculation that enables rapid repolarisation.[30] At the other extreme, in models of focal cerebral ischaemia, spontaneous depolarisations arise in isoelectric cortex or induce prolonged periods of isoelectric depression, and are accompanied by the inverse neurovascular response, vasoconstriction, which is termed cortical spreading ischaemia.[11,16,31] This inverse vasoreactivity initiates a vicious cycle of persisting depolarisation (up to ≥2 h) and metabolic substrate deficit that results in cortical necrosis. Since electrical silence is seen in the ischaemic penumbra,[26,27] isoelectricity was used as the criterion to distinguish these events in patients. The large odds ratio for unfavourable outcome (7·58) and 60% mortality for patients with ISD confirm that these events have a severe prognosis. The odds ratio for CSD was more modest (1·56) and not significant, and these individuals had mortality rates similar to those with no depolarisations. Because a continuous range of spreading depolarisations takes place in patients with TBI,[20] more refined classification methods that capture the graded severity of spreading depolarisations should increase prognostic sensitivity and better define the relation of depolarisation burden to outcome.[10]

Detection of spreading depolarisations currently requires that an electrode strip be placed on the brain surface, restricting the study population to patients who undergo craniotomy and raising the question of generalisability of our findings to a non-surgical population. Within the range of TBI,[32] most patients in our cohort had one of two types of injury: focal contusions and subdural haematomas. Subarachnoid haemorrhage was present in 83 (81%) individuals, and in a separate analysis, we noted that the only measure of CT ab-normalities associated with spreading depolarisations is the degree of subarachnoid haemorrhage (unpublished data). Since subarachnoid haemorrhage is common in moderate-to-severe TBI—arising, for example, in 53% of a broad sample of 1201 patients from the trials of tirilazad[33]—our findings might also apply to patients with contusions or subdural haematomas who are not treated surgically. On the other hand, the 9% of outcome variance that could be explained by the prognostic score is perhaps lower than expected and could suggest that predictive models behave differently in our highly selected cohort. For instance, patients with primarily diffuse swelling, diffuse axonal injury, or epidural haematomas were not well represented in our study; their mechanisms of tissue damage could differ and their exclusion might affect the performance of predictive models. Thus, in view of the few patients and low percentage of variance explained by the prognostic score, validation of our findings in additional cohorts is important. These cohorts would preferably include a non-surgical population, if less invasive methods such as scalp electroencephalography or near-infrared spectroscopy can be developed for monitoring of spreading depolarisations.

A further methodological limitation is the limited spatial sampling of one electrode strip, which is also a difficulty for most neuromonitoring technologies, such as partial pressure of brain tissue oxygen ($PbtO_2$). By contrast with monitoring of $PbtO_2$, however, electrode strips measure activity over a greater distance of 5 cm and, because spreading depolarisations propagate, probably capture spreading depolarisation incidence from an even broader territory. Location of strip placement also varies, owing to the variable anatomy of TBI, and this could affect detection of ISD and CSD. However, any variance attributable to these limitations would bias our data towards false negatives, which strengthens the interpretation of associations seen in our fairly small sample of 103 patients.

In summary, our results show the relevance of a novel pathological mechanism for monitoring and therapeutic targeting of patients with TBI. If our results are validated in additional cohorts, monitoring of spreading depolarisations could be used to guide treatment decisions in future clinical trials, in line with the recommendations to individualise clinical management and use mechanistic targets.[34] Several pharmacological and physiological therapeutic options for spreading depolarisations could exist.[10,11] A non-invasive method for monitoring of spreading depolarisations would accelerate efforts to validate these findings and investigate their application.

Panel: Research in context

Systematic review

We searched PubMed between January, 1970, and September, 2011, to identify studies in which spreading depolarisations were measured and outcomes after traumatic brain injury were reported, with the search terms "(spreading depolarisations OR spreading depression) AND (head injury OR traumatic brain injury)". Of seven studies identified, five reported five trauma patients or fewer, and two studies reported subgroups from the present cohort.[9,20] In the largest series of 53 patients, univariate analysis showed an association of depolarisation category with outcome. Expanding the disease search to "intracerebral hemorrhage OR subarachnoid hemorrhage OR stroke" returned two studies in which outcomes were analysed. Ten patients with large middle cerebral artery strokes all had unfavourable outcomes (extended Glasgow outcome score 1–4) and eight of ten had isoelectric spreading depolarisations (termed peri-infarct depolarisations in the report).[23] In 18 patients with aneurysmal subarachnoid haemorrhage, depolarisations with depression periods longer than 10 min had worse outcomes at discharge and the rate of depolarisation was higher in patients who developed delayed cerebral ischaemia.[15]

Interpretation

As far as we know, our study is the largest to date that examines the relation of spreading depolarisations to clinical outcome in acute brain injury and controls for baseline prognostic factors. Our results provide strong evidence that spreading depolarisations are independently associated with unfavourable outcomes after head trauma. Isoelectric spreading depolarisations in particular have poor prognosis, as also noted in stroke patients,[23] whereas evidence remains inconclusive for the effects of spreading depolarisations that depress ongoing spontaneous activity.

Articles

References

1 Maas AI, Stocchetti N, Bullock R. Moderate and severe traumatic brain injury in adults. *Lancet Neurol* 2008; **7:** 728–41.

2 Marmarou A, Lu J, Butcher I, et al. IMPACT database of traumatic brain injury: design and description. *J Neurotrauma* 2007; **24:** 239–50.

3 Roozenbeek B, Maas AI, Lingsma HF, et al. Baseline characteristics and statistical power in randomized controlled trials: selection, prognostic targeting, or covariate adjustment? *Crit Care Med* 2009; **37:** 2683–90.

4 Steyerberg EW, Mushkudiani N, Perel P, et al. Predicting outcome after traumatic brain injury: development and international validation of prognostic scores based on admission characteristics. *PLoS Med* 2008; **5:** e165.

5 Murray GD, Butcher I, McHugh GS, et al. Multivariable prognostic analysis in traumatic brain injury: results from the IMPACT study. *J Neurotrauma* 2007; **24:** 329–37.

6 Maas AI, Steyerberg EW, Marmarou A, et al. IMPACT recommendations for improving the design and analysis of clinical trials in moderate to severe traumatic brain injury. *Neurotherapeutics* 2010; **7:** 127–34.

7 Strong AJ, Fabricius M, Boutelle MG, et al. Spreading and synchronous depressions of cortical activity in acutely injured human brain. *Stroke* 2002; **33:** 2738–43.

8 Fabricius M, Fuhr S, Bhatia R, et al. Cortical spreading depression and peri-infarct depolarization in acutely injured human cerebral cortex. *Brain* 2006; **129:** 778–90.

9 Hartings JA, Strong AJ, Fabricius M, et al. Spreading depolarizations and late secondary insults after traumatic brain injury. *J Neurotrauma* 2009; **26:** 1857–66.

10 Lauritzen M, Dreier JP, Fabricius M, Hartings JA, Graf R, Strong AJ. Clinical relevance of cortical spreading depression in neurological disorders: migraine, malignant stroke, subarachnoid and intracranial hemorrhage, and traumatic brain injury. *J Cereb Blood Flow Metab* 2011; **31:** 17–35.

11 Dreier JP. The role of spreading depression, spreading depolarization and spreading ischemia in neurological disease. *Nat Med* 2011; **17:** 439–47.

12 Leão AAP. Spreading depression of activity in the cerebral cortex. *J Neurophysiol* 1944; **7:** 359–90.

13 Somjen GG. Mechanisms of spreading depression and hypoxic spreading depression-like depolarization. *Physiol Rev* 2001; **81:** 1065–96.

14 Hossmann KA. Periinfarct depolarizations. *Cerebrovasc Brain Metab Rev* 1996; **8:** 195–208.

15 Dreier JP, Woitzik J, Fabricius M, et al. Delayed ischaemic neurological deficits after subarachnoid haemorrhage are associated with clusters of spreading depolarizations. *Brain* 2006; **129:** 3224–37.

16 Dreier JP, Major S, Manning A, et al. Cortical spreading ischaemia is a novel process involved in ischaemic damage in patients with aneurysmal subarachnoid haemorrhage. *Brain* 2009; **132:** 1866–81.

17 Nakamura H, Strong AJ, Dohmen C, et al. Spreading depolarizations cycle around and enlarge focal ischaemic brain lesions. *Brain* 2010; **133:** 1994–2006.

18 Bosche B, Graf R, Ernestus RI, et al. Recurrent spreading depolarizations after subarachnoid hemorrhage decreases oxygen availability in human cerebral cortex. *Ann Neurol* 2010; **67:** 607–17.

19 Sakowitz OW, Kiening KL, Krajewski KL, et al. Preliminary evidence that ketamine inhibits spreading depolarizations in acute human brain injury. *Stroke* 2009; **40:** e519–22.

20 Hartings JA, Watanabe T, Bullock MR, et al. Spreading depolarizations have prolonged direct current shifts and are associated with poor outcome in brain trauma. *Brain* 2011; **134:** 1529–40.

21 Hukkelhoven CW, Steyerberg EW, Habbema JD, et al. Predicting outcome after traumatic brain injury: development and validation of a prognostic score based on admission characteristics. *J Neurotrauma* 2005; **22:** 1025–39.

22 Brain Trauma Foundation, American Association of Neurological Surgeons, Congress of Neurological Surgeons. Guidelines for the management of severe traumatic brain injury. *J Neurotrauma* 2007; **24** (suppl 1): S1–106.

23 Dohmen C, Sakowitz OW, Fabricius M, et al. Spreading depolarizations occur in human ischemic stroke with high incidence. *Ann Neurol* 2008; **63:** 720–28.

24 Busch E, Gyngell ML, Eis M, HoehnBerlage M, Hossmann KA. Potassium-induced cortical spreading depressions during focal cerebral ischemia in rats: contribution to lesion growth assessed by diffusion-weighted NMR and biochemical imaging. *J Cereb Blood Flow Metab* 1996; **16:** 1090–99.

25 Back T, Ginsberg MD, Dietrich WD, Watson BD. Induction of spreading depression in the ischemic hemisphere following experimental middle cerebral artery occlusion: effect on infarct morphology. *J Cereb Blood Flow Metab* 1996; **16:** 202–13.

26 Astrup J, Symon L, Branston NM, Lassen NA. Cortical evoked potential and extracellular K+ and H+ at critical levels of brain ischemia. *Stroke* 1977; **8:** 51–57.

27 Astrup J, Siesjo BK, Symon L. Thresholds in cerebral ischemia: the ischemic penumbra. *Stroke* 1981; **12:** 723–25.

28 McHugh GS, Butcher I, Steyerberg EW, et al. A simulation study evaluating approaches to the analysis of ordinal outcome data in randomized controlled trials in traumatic brain injury: results from the IMPACT Project. *Clin Trials* 2010; **7:** 44–57.

29 Jones PA, Andrews PJD, Midgley S, et al. Measuring the burden of secondary insults in head-injured patients during intensive care. *J Neurosurg Anesthesiol* 1994; **6:** 4–14.

30 Piilgaard H, Lauritzen M. Persistent increase in oxygen consumption and impaired neurovascular coupling after spreading depression in rat neocortex. *J Cereb Blood Flow Metab* 2009; **29:** 1517–27.

31 Strong AJ, Anderson PJ, Watts HR, et al. Peri-infarct depolarizations lead to loss of perfusion in ischaemic gyrencephalic cerebral cortex. *Brain* 2007; **130:** 995–1008.

32 Saatman KE, Duhaime A-C, Bullock R, et al. Classification of traumatic brain injury for targeted therapies. *J Neurotrauma* 2008; **25:** 719–38.

33 Maas AIR, Hukkelhoven CWMP, Marshall LF, Steyerberg EW. Prediction of outcome in traumatic brain injury with computed tomographic characteristics: a comparison between the computed tomographic classification and combinations of computed tomographic predictors. *Neurosurgery* 2005; **57:** 1173–82.

34 Maas AIR, Menon DK, Lingsma HF, Pineda JA, Sandel ME, Manley GT. Re-orientation of clinical research in traumatic brain injury: report of a international workshop on comparative effectiveness research. *J Neurotrauma* 2011; published online Aug 29. DOI:10.1089/neu.2010.1599.

questionable, and the statistical significance for TMS could be spurious.

The data from this trial suggest that, contrary to what the investigators expected, the benefits of pridopidine are more pronounced for some items of the UHDRS–TMS that were not included in the mMS (eye movements and dystonia) than for mMS items. A second trial, although not yet published in a peer-reviewed journal, has found similar results, suggesting that pridopidine-related benefits, although subtle, are real.[5] Analysis of individual items within the UHDRS–TMS in the MermaiHD study also suggests that pridopidine might benefit features of Huntington's disease for which there are currently no treatments (eye movements, hand coordination, dystonia, and gait or balance problems). Future trials will be needed to confirm these potential effects, and to investigate whether functional benefits accompany the motor improvements. Additionally, since the 90 mg dose was well tolerated, perhaps higher doses could be tried in future trials.

A well tolerated drug that produces even small benefits for patients with Huntington's disease would be a very welcome addition to the currently available treatments for this debilitating disorder. The MermaiHD trial raises the hope that dopamine-stabilising agents will be useful for treating the symptoms of Huntington's disease. Although this trial does not provide conclusive evidence that pridopidine is an effective treatment for Huntington's disease, it is a welcome step in the right direction.

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1 Huntington G. On chorea. *Med Surg Rep* 1872; **26:** 320–21.
2 Peavy GM, Jacobson MW, Goldstein JL, et al. Cognitive and functional decline in Huntington's disease: dementia criteria revisited. *Mov Disord* 2010; **25:** 1163–69.
3 Marder K, Zhao H, Myers RH, et al. Rate of functional decline in Huntington's disease. *Neurology* 2000; **54:** 452–58.
4 Garcia de Yebenes J, Landwehrmeyer B, Squitieri F, et al, for the MermaiHD study investigators. Pridopidine for the treatment of motor function in patients with Huntington's disease: a phase 3, randomised, placebo-controlled trial. *Lancet Neurol* 2011. Published online Nov 8. DOI:10.1016/S1474-4422(11)70233-2
5 The Huntington Study Group HART Investigators, McGarry A, Kieburtz K, Rembratt A, Tedroff J. A randomized, double-blind placebo-controlled trial of ACR16 in Huntington's disease. *Neurotherapeutics* 2011; **8:** 135.

Traumatic brain injury: prognostic implications of cortical electrical disturbances

Traumatic brain injury (TBI) can be exacerbated by various mechanisms. Some are well known, such as tissue hypoxia due to arterial hypotension or raised intracranial pressure, but some are more difficult to identify. Subdural electrocorticographic recording, through a strip of electrodes inserted after a neurosurgical operation, has shown waves of depolarisation in the cortex of patients with acute brain damage of various causes.[1] The occurrence of these waves is associated with impairment of perfusion and energy metabolism, carrying the risk that primary brain injury might expand and worsen over time. The consequences of cortical depolarisation, which have been well investigated in animal experiments, have also been detected in patients. Results from a study[2] that used online microdialysis have shown that multiple cortical depolarisations lead to local depletion of brain glucose and energy crisis. This could compromise cellular repolarisation and tissue integrity. On the basis of these findings, cortical depolarisation is not simply an electrical disturbance of a suffering cortex; on the contrary, it could cause a clinically significant metabolic disorder, thus worsening outcome.

In this issue of *The Lancet Neurology*, Jed Hartings and colleagues[3] analyse the prognostic implications of cortical depolarisation in 103 patients with TBI. The main conclusions of the study are that cortical depolarisation is frequent and is independently associated with worse outcome 6 months after injury. This result confirms, with a more elaborate multivariable analysis, the association with worse outcome previously reported by the same group in 53 patients.[4] Should, therefore, cortical depolarisation be more actively sought in TBI, and become a target of potential treatments? This

Comment

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hypothesis is worth investigating, but several points require particular work.

First, we still do not know how frequent cortical depolarisation is in TBI. For electrocorticography to be possible a neurosurgical intervention is necessary, and data on cortical depolarisation have been obtained only in surgical cases. Whereas 60% of those cases might exhibit cortical depolarisation, we have no data on most patients with TBI, who do not undergo surgery. Moreover, the patients included in this study[3] have especially severe injuries, since 45% of patients underwent surgery because of delayed deterioration.

Second, electrocorticography explores a portion of the brain that has been injured, in which contusions have been resected or expanding masses have compressed the tissue. The extent to which the disturbances recorded in these areas represent what happens in other portions of the brain, and how these disturbances contribute to overall disability, remains to be clarified.

Third, not all cortical depolarisations have the same prognostic value, and the effects of different types need to be further investigated. Although a strong association with worse outcome was shown for isoelectric spreading depolarisation (ISD), no significant association was reported for cortical spreading depression (CSD) in the present study.[3]

Fourth, the real burden of cortical depolarisation, and more specifically ISD, on outcome remains unclear. Although the association of ISD with unfavorable outcome seems clear, statistical analysis does not rule out the possibility that ISD was detected in patients with especially severe injuries, which could also contribute to the worse outcome. Cortical depolarisation has been associated with low perfusion and high temperature after TBI.[5] Unfortunately, data on concurrent changes in intracranial pressure, cerebral perfusion pressure, tissue oxygenation, and fever are not reported, so the contributions of these factors to cortical depolarisation and outcome remain unknown.

Finally, the inclusion of depolarisation category (none, ISD, or CSD only) in the prognostic model increased the explained variance in outcome (Nagelkerke R^2) from 9% to 22%.[3] The Nagelkerke R^2, an index of the prognostic strength of the model, suggests a relevant role for cortical depolarisation. At the same time, this variable shows that the other covariates account for only 9% of the variance, perhaps reflecting the fact that all the selected predictors were admission variables (eg, age, pupilary reactivity, and motor score). Inclusion of other parameters (eg, intracranial pressure, cerebral perfusion pressure, tissue oxygenation, and temperature) in the model would have been interesting. In summary, depolarisations seem to contribute strongly to prognosis, but in a model in which 78% of the variance of the outcome is not explained.

Despite these limitations, we know that depolarisations have been detected in a significant proportion of patients with surgically treated TBI in repeated studies. Previous evidence[1,2] shows that cortical depolarisations might contribute to tissue damage. Data from this study[3] prove that depolarisations are associated with worse outcome in a fairly large cohort. What was once an esoteric finding now seems to be more relevant, and worthy of being monitored, better understood, and possibly a target for intervention.

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1   Lauritzen M, Dreier JP, Fabricius M, Hartings JA, Graf R, Strong AJ. Clinical relevance of cortical spreading depression in neurological disorders: migraine, malignant stroke, subarachnoid and intracranial hemorrhage, and traumatic brain injury. *J Cereb Blood Flow Metab* 2011; **31:** 17–35.

2   Feuerstein D, Manning A, Hashemi P, et al. Dynamic metabolic response to multiple spreading depolarizations in patients with acute brain injury: an online microdialysis study. *J Cereb Blood Flow Metab* 2010; **30:** 1343–55.

3   Hartings JA, Bullock MR, Okonkwo DO, et al. Spreading depolarisations and unfavourable outcome in traumatic brain injury: a prospective, observational study. *Lancet Neurol* 2011; published online Nov 3. DOI:10.1016/S1474-4422(11)70243-5.

4   Hartings JA, Watanabe T, Bullock MR, et al. Spreading depolarizations have prolonged direct current shifts and are associated with poor outcome in brain trauma. *Brain* 2011; **134:** 1529–40.

5   Hartings JA, Strong AJ, Fabricius M, et al. Spreading depolarizations and late secondary insults after traumatic brain injury. *J Neurotrauma* 2009; **26:** 1857–66.

Predictors of multiple sclerosis in children and implications for early initiation of treatment

Recent major advances in the care of children and adolescents with multiple sclerosis (MS) relate to the understanding of the natural history of the disease and to the effect of disease-modifying therapy in this group of patients. These advances highlight the importance of being able to predict which children and adolescents are at greatest risk of developing MS. Leonard Verhey and colleagues[1] report the results of an MRI study in this issue of *The Lancet Neurology* that aims to address this important question.

As with the study reported in this issue, the recent advances on natural history and the effect of disease-modifying therapy resulted from large epidemiological studies following children from their first CNS demyelinating event. In relation to the natural history of disease, we know now that the initial demyelinating events can be clinically diverse, including occurrence of acute disseminated encephalomyelitis (ADEM), especially in children younger than 10 years; the rate of relapses is high during the first years of disease and the motor disability is a late event, while cognitive impairment is probably more frequent than motor disability.[2]

With regard to the effect of disease-modifying therapies (mostly interferons and glatiramer acetate), we know now that they are safe in children younger than 12 years, and most probably effective at reducing the initial clinical relapse rate and more effective if initiated early after disease onset.[3] However, availability of new drugs, beyond interferons and glatiramer acetate, increases our need for knowledge about the natural history of MS in children. This new information is necessary for the development of paediatric investigation plans, which pharmaceutical companies must develop and that should be approved by regulatory agencies before licensing a new medicine for children. Early initiation of treatment requires recognition of MS at the time of first demyelinating event with a high level of confidence in the absence of a much needed but elusive biological marker; elaboration of a paediatric investigation plan for a new compound requires surrogate markers of disease development. Elaboration of specific plans for paediatric patients

Spreading depolarisations and traumatic brain injury: time course and mechanisms

In a recent research article in *The Lancet Neurology*,[1] Jed A Hartings and colleagues reported their findings of electrocorticography in 103 patients requiring intensive care and surgery for traumatic brain injury (TBI). They found an association between the presence and type of spreading depolarisations during acute ictus and survival and functional outcome 6 months later. The investigators suggest that, after further clinical studies, individualised care could include monitoring and treatment of spreading depolarisations. Two issues arise from this excellent report.

Patients were recruited into the study within 7 days of TBI and electrocorticography was done for the duration of other clinical invasive monitoring, or to a maximum of 7 days. Electrocorticography monitoring continued for 67 h (IQR 40–102) but, theoretically, a patient could have been monitored as late as 14 days after injury. First, what was the time course of the spreading depolarisations? Were cortical spreading depolarisations clustered in the early phase after injury, as in experimental middle cerebral artery occlusion?[2] Alternatively, were isoelectric spreading depolarisations predominantly preterminal phenomena? In this regard, I note that isoelectric spreading depolarisations occurred in 12 of 28 patients that died compared with their occurrence in eight of 75 patients that survived (43% vs 11%, SD 3·6 difference in the hypothesis test for proportions, p<0·001).

In 1999, Morris and colleagues[3] reported the failure of two phase 3 clinical trials of a competitive NMDA-receptor antagonist (Selfotel, CGS 19755) in the treatment of severe TBI. In the international arm of the trial, there was a subgroup assessment of patients who underwent operation for mass lesions (which was similar to the types of operations and patients in Hartings and colleagues' study[1]). On inspection of these data—in view of the limitations of small sample size and a quick post-hoc analysis using information from the tables—the use of intravenous Selfotel (5 mg/kg once a day) for 4 days did not seem to reduce the proportion of patients dying (more deaths in the Selfotel group, 31% vs 20%, SD 1·5, p>0·05). Also, in the Selfotel group, there was a smaller proportion of favourable outcomes (42% vs 67%, SD 2·9 difference, p<0·01) than in the placebo group.

The electrophysiological and ionic transients of cortical spreading depolarisations are NMDA-receptor mediated, and inhibition can be induced by agents acting at the NR2B, glycine, or non-competitive binding sites.[4–7] The Selfotel trials[3] can be considered a proof of concept or even a test of the hypothesis proposed by Hartings and colleagues' in their conclusion—short-term inhibition of a cortical NMDA receptor-mediated process in severe TBI cases requiring neurosurgery fails to improve outcome. Therefore, the second point of discussion that I would like to raise is the type of tests of their hypothesis they envisage. For example, they could take the form of a longer duration of interventional therapy, up to 14 days—for which it would be important to know more about the clustering and preterminal information. Alternatively, tests could involve the use of NMDA-receptor antagonists that have both a neuronal and oligodendrocyte profile.[8,9] Such protection of white matter injury in severe TBI would indeed be inkeeping with outcome long after such injury.[10]

I declare that I have no conflicts of interest.

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1 Hartings JA, Bullock MR, Okonkwo DO, et al. Spreading depolarisations and outcome after traumatic brain injury: a prospective observational study. *Lancet Neurol* 2011; **10:** 1058–64.
2 Hartings JA, Rolli ML, Lu XC, Tortella FC. Delayed secondary phase of peri-infarct depolarizations after focal cerebral ischemia: relation to infarct growth and neuroprotection. *J Neurosci* 2003; **27:** 11602–10.
3 Morris GF, Bullock R, Marshall SB, et al. Failure of the competitive N-methyl-D-aspartate antagonist Selfotel (CGS 19755) in the treatment of severe head injury: results of two phase III clinical trials. *J Neurosurg* 1999; **91:** 737–43.
4 McLachlan RS. Suppression of spreading depression of Leao in neocortex by an N-methyl-D-aspartate receptor antagonist. *Can J Neurol Sci* 1992; **19:** 487–91.
5 Nellgard B, Wieloch T. NMDA-receptor blockers but not NBQX, an AMPA-receptor antagonist, inhibit spreading depression in the rat brain. *Acta Physiol Scand* 1992; **146:** 497–503.
6 Obrenovitch TP, Zilkha E. Inhibition of cortical spreading depression by L-701,324, a novel antagonist at the glycine site of the N-methyl-D-aspartate receptor complex. *Br J Pharmacol* 1996; **117:** 931–37.
7 Somjen GG. Mechanisms of spreading depression and hypoxic spreading depression-like depolarization. *Physiol Rev* 2001; **81:** 1065–96.
8 Bakiri Y, Hamilton NB, Karadottir R, Attwell D. Testing NMDA receptor block as a therapeutic strategy for reducing ischemic damage to CNS white matter. *Glia* 2008; **56:** 233–40.
9 Giacino JT, Whyte J, Bagiella E, et al. Placebo-controlled trial of amantadine for severe traumatic brain injury. *N Engl J Med* 2012; **366:** 819–26.
10 Tasker RC, Westland AG, White DK, Williams GB. Corpus callosum and inferior forebrain white matter microstructure are related to functional outcome from raised intracranial pressure in child traumatic brain injury. *Dev Neurosci* 2010; **32:** 374–84.

Authors' reply

We appreciate Robert C Tasker's interest in our report[1] and thank him for his interesting questions. His concerns relate to the temporal patterning of cortical spreading depolarisations and to the implications for design of therapeutic trials.

In the figure we present a different display of the same data reported in our study to show the timing of electrocorticography and the spreading depolarisations. Recordings were obtained mainly in the first week
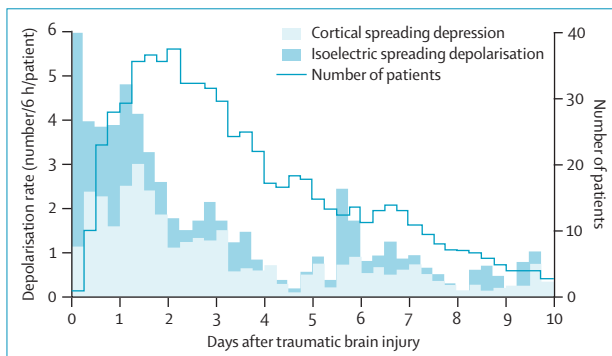
Figure: Time course of depolarisations after traumatic brain injury

The total number of depolarisations in each 6 h bin was divided by the number of patients monitored with electrocorticography for the corresponding bin to calculate depolarisation rates than can be compared across time. This eliminates bias in the raw depolarisation counts created by the varying numbers of patients monitored through time. Rates for isoelectric spreading depolarisations and cortical spreading depression were computed separately and are stacked to show overall depolarisation rate. Non-integer values in the patient numbers result from start or termination of recordings within a 6-h time bin (eg, 4-h recording=0·67 patients). Data are derived from the 58 patients with depolarisations reported in our study.[1]

after TBI. Depolarisations occurring in electrically silent cortex (or isoelectric spreading depolarisations [ISD]) accounted for 36% of all events and showed a similar time course to depolarisations with depression of spontaneous cortical activity (cortical spreading depression): a maximal activity in the initial 48 h, followed by a decline and then a delayed secondary peak on days 6–8.[2,3] Thus, the association of ISD with poor outcome is not because ISD occur only as preterminal events associated with global brain death. The predominance of ISD within the initial days rather suggests that, as in ischaemic stroke, ISD are associated with acute lesion development. The association with mortality might be both correlative and causative, since isoelectricity could result from ischaemia or impaired neurovascular coupling,[4] and depolarisations cause cell death under these conditions, probably through Ca^{2+}-mediated pathways.

Assuming that NMDA-receptor antagonism inhibits all depolarisations, which is unlikely,[5] does the failure of the Selfotel trials[6] provide evidence against depolarisations as a promising therapeutic target? We don't think so. First, if there was a beneficial effect from

blocking spreading depolarisations, it would occur only in the 56% of patients with depolarisations, or perhaps just the 20% with ISD. Thus any effect would be greatly diluted by broad inclusion, even within the surgical subgroup. Second, it is questionable whether sufficient brain levels of Selfotel, a competitive antagonist, were achieved to compete with high glutamate concentrations (100–200 times more than normal) for receptor binding; there was no confirmation of intended effect on the target mechanism. In a subsequent severe TBI trial with the non-competitive NR2B-selective antagonist traxoprodil, improvements in outcome up to 11·8% were reported in some subgroups.[7] Third, the Selfotel trials were stopped in part for safety concerns in a concomitant stroke trial, so that sufficient statistical power was never achieved.

We envision electrocorticography as another method in the armamentarium of neuromonitoring. Just as mannitol is given only to patients with elevated intracranial pressure, a therapy to block spreading depolarisations would be administered only to patients that have them and for as long as they persist. This will confirm that the pathological processes targeted by therapeutics are active in individual patients. The advantages of this approach are selective inclusion, mechanistic targeting, and, not least, assessment of the effect of therapy on the targeted mechanism. Such a design differs dramatically from the failed trials in the past, but rather builds towards personalised treatment of this heterogeneous disease. More work is needed to determine which depolarisations should be targeted, which therapies are most effective, and to ensure that interventions do not interfere with repair processes.[8,9]

We declare that we have no conflicts of interest.

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1 Hartings JA, Bullock MR, Okonkwo DO, et al. Spreading depolarisations and outcome after traumatic brain injury: a prospective observational study. *Lancet Neurol* 2011; **10:** 1058–64.

2 Hartings JA, Strong AJ, Fabricius M, et al. Spreading depolarizations and late secondary insults after traumatic brain injury. *J Neurotrauma* 2009; **26:** 1857–66.

3 Dreier JP, Major S, Pannek HW, et al. Spreading convulsions, spreading depolarization and epileptogenesis in human cerebral cortex. *Brain* 2011; **135:** 259–75.

4 Dreier JP, Major S, Manning A, et al. Cortical spreading ischaemia is a novel process involved in ischaemic damage in patients with aneurysmal subarachnoid haemorrhage. *Brain* 2009; **132:** 1866–81.

5 Lauritzen M, Hansen AJ. The effect of glutamate receptor blockade on anoxic depolarization and cortical spreading depression. *J Cereb Blood Flow Metab* 1992; **12:** 223–29.

6 Morris GF, Bullock R, Marshall SB, Marmarou A, Maas A, Marshall LF. Failure of the competitive N-methyl-D-aspartate antagonist Selfotel (CGS 19755) in the treatment of severe head injury: results of two phase III clinical trials. The Selfotel Investigators. *J Neurosurg* 1999; **91:** 737–43.

7 Yurkewicz L, Weaver J, Bullock MR, Marshall LF. The effect of the selective NMDA receptor antagonist traxoprodil in the treatment of traumatic brain injury. *J Neurotrauma* 2005; **22:** 1428–43.

8 Lo EH. A new penumbra: transitioning from injury into repair after stroke. *Nat Med* 2008; **14:** 497–500.

9 Ikonomidou C, Turski L. Why did NMDA receptor antagonists fail clinical trials for stroke and traumatic brain injury? *Lancet Neurol* 2002; **1:** 383–86.

Epilepsy nurse specialists are a vital resource

I write in response to Mario Christodoulou's feature[1] about threats to the vital support provided by neurological nurse specialists in the UK. As the chief executive of Young Epilepsy, a UK charity working to improve health and education services for young people with epilepsy, I wholeheartedly support the arguments presented in the report, which highlighted the individualised care and cost benefits that specialist nurses bring to the health-care team.

The UK National Institute for Health and Clinical Excellence clinical

BRAIN
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Effect of analgesics and sedatives on the occurrence of spreading depolarizations accompanying acute brain injury

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Spreading depolarizations are waves of mass neuronal and glial depolarization that propagate across the injured human cortex. They can occur with depression of neuronal activity as spreading depressions or isoelectric spreading depolarizations on a background of absent or minimal electroencephalogram activity. Spreading depolarizations are characterized by the loss of neuronal ion homeostasis and are believed to damage functional neurons, leading to neuronal necrosis or neurological degeneration and poor outcome. Analgesics and sedatives influence activity-dependent neuronal ion homeostasis and therefore represent potential modulators of spreading depolarizations. In this exploratory retrospective international multicentre analysis, we investigated the influence of midazolam, propofol, fentanyl, sufentanil, ketamine and morphine on the occurrence of spreading depolarizations in 115 brain-injured patients. A surface electrode strip was placed on the cortex, and continuous electrocorticographical recordings were obtained. We used multivariable binary logistic regression to quantify associations between the investigated drugs and the hours of electrocorticographical recordings with and without spreading depolarizations or clusters of spreading depolarizations. We found that administration of ketamine was associated with a reduction of spreading depolarizations and spreading depolarization clusters ($P < 0.05$). Midazolam anaesthesia, in contrast, was associated with an increased number of spreading depolarization clusters ($P < 0.05$). By using a univariate odds ratio analysis, we also found a significant

association between ketamine administration and reduced occurrence of isoelectric spreading depolarizations in patients suffering from traumatic brain injury, subarachnoid haemorrhage and malignant hemispheric stroke ($P < 0.05$). Our findings suggest that ketamine—or another *N*-methyl-D-aspartate receptor antagonist—may represent a viable treatment for patients at risk for spreading depolarizations. This hypothesis will be tested in a prospective study.

Introduction

Spreading depolarizations are characterized by the abrupt, near-complete breakdown of ion gradients across cellular membranes, the sustained depolarization of neurons and glia, loss of brain electrical activity and neuronal swelling. This term describes the full spectrum of such cortical waves, from terminal depolarizations, characteristic of severe anoxia and ischaemia, to the short-lasting depolarizations assumed to underlie migraine aura (Strong *et al.*, 2002; Dreier *et al.*, 2006; Dreier, 2011). Electrocorticographically, spreading depolarizations are characterized by a large and slow potential change (Canals *et al.*, 2005). When neuronal metabolism is largely intact before the onset of spreading depolarizations, it leads to a depression of neuronal activity (Strong *et al.*, 2002), because the accompanying sustained neuronal depolarization lies above the inactivation threshold for action potential-generating ion channels (Karger *et al.*, 2002). On the other hand, when neuronal metabolism is disturbed before spreading depolarization onset, neuronal activity has already ceased and spreading depolarizations cannot trigger a further depression of activity. Under these conditions, spreading depolarizations are termed as isoelectric spreading depolarizations to distinguish them from spreading depressions (Hartings *et al.*, 2011*a*). Temporally clustered spreading depolarizations are associated with the development of new infarcts after aneurysmal subarachnoid haemorrhage and with poorer outcome after subarachnoid haemorrhage and traumatic brain injury (Dreier *et al.*, 2006; Dreier, 2011; Hartings *et al.*, 2011*a*). Moreover, they are frequently observed in malignant hemispheric stroke (Dohmen *et al.*, 2008). Thus, the occurrence of isoelectric spreading depolarizations is probably associated with a poorer prognosis than the occurrence of spreading depressions (Fabricius *et al.*, 2006; Hartings *et al.*, 2012).

Whether spreading depolarizations can be modulated pharmacologically in the human brain is largely unknown (Lauritzen *et al.*, 2011; Dreier, 2011). However, our preliminary findings suggest that large doses of analgesic and sedative drugs—such as are administered on intensive care units—may influence the occurrence of spreading depolarizations (Sakowitz *et al.*, 2009). Interestingly, most recordings of spreading depolarizations are made under the influence of such analgesic and sedative drugs. These include enhancers of γ-aminobutyric acid (GABA) receptor action such as benzodiazepines and barbiturates, opioid receptor agonists and the *N*-methyl-D-aspartate (NMDA) receptor blocker, ketamine. These drugs target receptors that regulate neuronal activity and synaptic transmission and may thereby alter susceptibility to and course of spreading depolarizations (Somjen, 2001).

Although the occurrence of spreading depolarizations may presumably be modulated by a broad range of molecular and physical entities, we chose in this study to focus solely on the influence mediated by a selected set of drugs commonly employed on intensive care units. The goal of our analyses was to identify the possible drug candidates for reducing the occurrence of spreading depolarizations in patients following acute brain injury.

Materials and methods

We received 115 sets of patient data from seven member centres of the international multicentre observational study group, Cooperative Study of Brain Injury Depolarizations (COSBID) (www.cosbid.org), in Europe and USA (Cologne, Cincinnati, Berlin, Heidelberg, London, Richmond and Pittsburgh). All procedures were carried out in accordance with ethical standards and were approved by local research ethics committees. After a decision for surgery (craniotomy) was made, patient or guardian consent was obtained according to the Declaration of Helsinki. For the analyses presented here, we collected prospective data including gender, age, disease group, time of ictus, time(s) of spreading depolarization events and outcome 6 months after surgery, which was quantified using the extended Glasgow Outcome Scale. These data belong to the COSBID basic data set and were obtained from the secure COSBID online database (www.swinklink. com/caboodle). In addition, all participating centres provided patient sedation logs. Because some centres recorded drug dose data in bedside patient charts by clock-hour, the highest resolution for our analysis was 1 h. Administered analgesics and sedatives included midazolam, flunitrazepam, thiopental, fentanyl, sufentanil, remifentanil, ketamine, propofol, γ-hydroxybutyric acid, morphine and clonidine. One centre also reported 17 bolus administrations of oxycodone. We focused on those drugs for which the total hours of electrocorticographic recordings exceeded 1000: midazolam, fentanyl, sufentanil, ketamine, propofol and morphine (Table 1). Flunitrazepam (80 h, $n = 2$ patients), thiopental (222 h, $n = 13$), remifentanil (200 h, $n = 9$), γ-hydroxybutyric acid (57 h, $n = 2$) and clonidine (180 h, $n = 17$) were not further investigated in this study. To normalize drug doses for further comparison, four quartiles were defined: $1 = {<}25\%$, $2 = 25\%$ quartile to below median, $3 =$ median to ${<}75\%$ and $4 = {\geqslant}75\%$ (Table 1).

Electrocorticographic recordings were initiated immediately after each craniotomy and placement of the subdural electrode strip, and on average 37 h ($\pm$41 h) after the initial stroke or trauma. In these data, we found that both the total number of recorded hours and the number of recorded hours with spreading depolarizations peaked on the third day after ictus (Fig. 1B). The time courses for drug administration, however, were different from both the distribution of total recorded hours and the recorded hours with spreading depolarizations

and also different for each individual drug under study. For example, although the number of recorded hours reached a peak 3 days after ictus (Fig. 1B), morphine administration peaked at 2 days after ictus, whereas ketamine administration did not reach a peak until 7 days after ictus (Fig. 1B).

The occurrence of multiple spreading depolarizations in close temporal proximity is typically referred to as a cluster. However, neither the precise number of spreading depolarizations within a cluster nor their frequency are yet defined. For this study, we defined a cluster as the smallest number of non-randomly distributed spreading depolarizations that could be temporally associated with a certainty of at least 99%. For all recorded hours in all patients, the probability that a spreading depolarization occurred in any given hour of recording was calculated as $(1850/15\,445) = 0.120 = 12.0\%$. It suggests that the probability for two spreading depolarizations to occur within the same hour or within two consecutive hours of recording was $(1850/15\,445)^2 = 0.014 = 1.4\%$ and that the probability that a sequence of at least three spreading depolarizations occurred within three consecutive hours of recording was $(1850/15\,445)^3 = 0.002 = 0.2\%$. Accordingly, the occurrence of at least three spreading depolarizations within three or fewer consecutive recording hours was considered to be a cluster.

Of the 115 data sets, 98 were complete (85%), 10 lacked an extended Glasgow Outcome Scale score, and the time of ictus was not indicated or unknown for eight data sets. For analysis of isoelectric spreading depolarizations, only data sets from patients with at least one isoelectric spreading depolarization were included.

Table 1 Investigated drugs with more than 1000 h of electrocorticographic recordings

Investigated drug	Recorded hours	Patients (n)	Median drug dose and quartiles (mg)		
			25%	Median	75%
Midazolam	5608	61	14.4	22.3	36
Fentanyl	5459	71	0.1	0.15	0.5
Propofol	3397	81	100	150	200
Sufentanil	1456	14	0.03	0.06	0.105
Ketamine	2168	26	125	200	300
Morphine	1646	30	6	8	10

Microsoft Excel® was used for data collection and generation of Pivot tables. We used an explorative approach to evaluate data. Univariate odds ratios and 95% confidence intervals were calculated using a custom routine. Statistical analyses were carried out using Graph Pad Prism® and IBM SPSS®. Statistical significance was set to $P < 0.05$.

Results

Surgical procedures in our group of patients were performed to treat traumatic brain injury, relieve compression after malignant hemispheric stroke, clip aneurysms and remove blood clots resulting from subarachnoid haemorrhage and intracerebral haemorrhage (Tables 2 and 3). Electrocorticographic recordings were obtained during postsurgical observation on the intensive care unit from a surface electrode strip placed on the cortex during each surgical procedure. We observed spreading depolarizations

Table 2 Patient characteristics overview

Patients	n = 115
Gender, n (%)	
Male	69 (60)
Female	46 (40)
Age (years)	range 18–79
	49.6 (±15)
Extended Glasgow	3 (1.5–5.5),
Outcome Scale (n = 105)	median (25–75% percentile)
Recorded ECoG	117.6 (±59.9)
hours and sedation logs	
Spreading depolarizations (n = 76)	24.3 (±27.7)
Recorded hours with spreading	17.6 (±18.2)
depolarizations (n = 76)	
Clustered spreading	13.9 (±13.5)
depolarizations (n = 47)	
Isoelectric spreading	9.77 (±13.6)
depolarizations (n = 13)	

Mean ± standard deviation, unless otherwise indicated. Outcome was assessed using an extended Glasgow Outcome Scale ranging from 1 (dead) to 8 (upper good recovery); ECoG = electrocorticography.
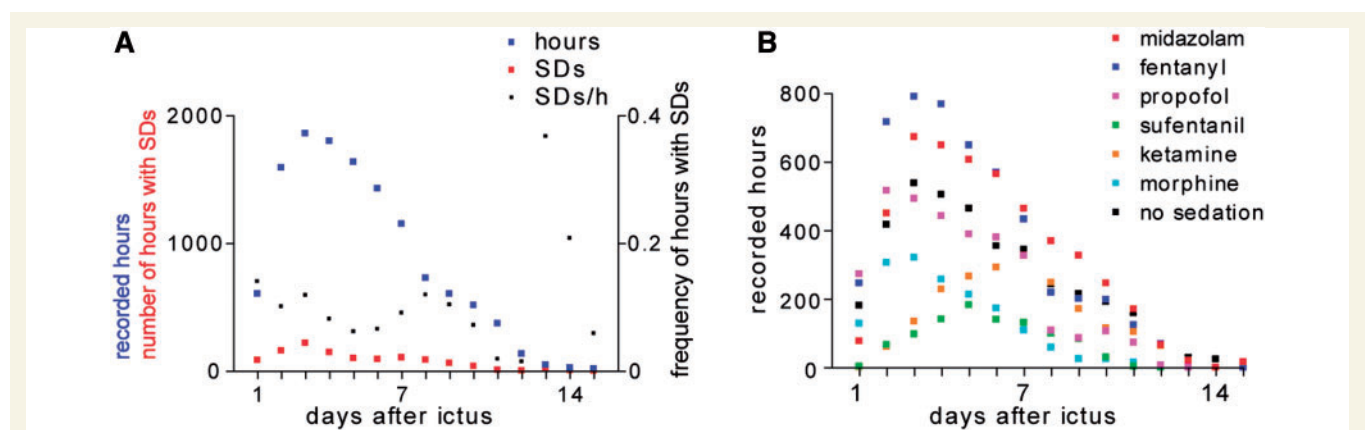
Figure 1 Recorded hours and administrations of sedatives and analgesics. (A) Total recorded hours of all patients and all observed spreading depolarizations (SDs). (B) The number of hours recorded for each drug depends on day of observation.

in 76 of 115 patients (66%). In our primary statistical analysis, frequencies of spreading depolarizations with and without administration of ketamine were analysed. More specifically, we compared paired relative frequencies of hours with spreading depolarizations with and without ketamine in individual patients. We

Table 3 Spreading depolarizations by type of brain injury

Disease group	Patients with spreading depolarizations (% incidence)	Mean spreading depolarizations frequency (SD/h ± standard deviation)
Traumatic brain injury ($n = 60$)	32 (53)	0.10 ($\pm$0.2)
SAH/ICH ($n = 31$)	24 (77)	0.10 ($\pm$0.1)
MHS ($n = 24$)	21 (88)	0.11 ($\pm$0.1)

ICH = intracerebral haemorrhage; MHS = malignant hemispheric stroke; SAH = subarachnoid haemorrhage; SD/h = spreading depolarizations per hour.

found that ketamine reduced the occurrence of spreading depolarizations (paired t-test $P < 0.05$, $n = 26$ patients). When evaluated using the same approach, no other drugs exhibited such differences (Fig. 2A). Binary multivariable logistic regression was used to investigate additional associations between drug administration and spreading depolarization occurrence. For this analysis, the drug dosage of 115 patients was normalized in quartiles (Table 1). All drugs for which >1000 h of electrocorticographic recordings were available were included in the model and were significantly associated with spreading depolarization occurrence (omnibus-test for χ^2; $P < 0.05$). The odds ratio for each of the six drugs was also determined (Table 4). According to these calculations, only ketamine administration was significantly associated with reduced occurrence of spreading depolarizations ($P < 0.05$).

Because choice of the analgesic and sedative drugs seemed to be influenced by the time passed since ictus, we evaluated spreading depolarization events and drug administration with respect to time (Fig. 1A and B). This analysis revealed that the reduction in
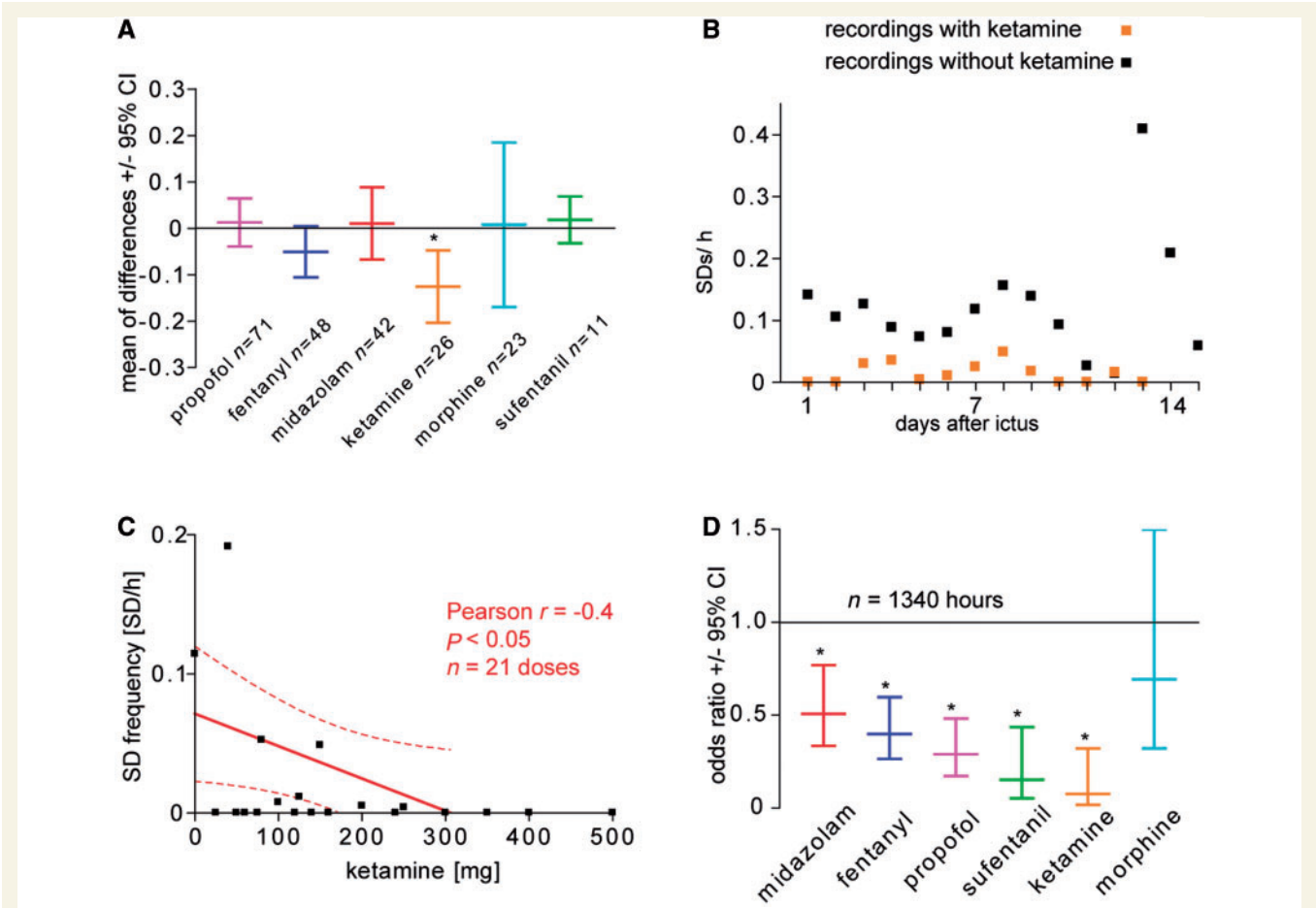
Figure 2 Ketamine markedly decreases the incidence of cortical spreading depolarizations in acutely brain-injured patients. (**A**) Ketamine administration within the individual patient reduced spreading depolarizations (SDs). Within-patient analysis of spreading depolarizations per hour of electrocorticographic recordings under drug influence and without analgesic or sedative drugs (n = number of included patients). (**B**) The inhibitory effect of ketamine on spreading depolarization occurrence is consistent across days and therefore independent of time since ictus. (**C**) Doses of ketamine were negatively correlated with spreading depolarization frequency. (**D**) Isoelectric spreading depolarization occurrence is influenced by analgesics and sedatives in a drug-dependent manner. *Significant at $P < 0.05$.

Table 4 Multivariate analysis of investigated analgesics and sedatives in 115 patients

	Odds ratio for spreading depolarization occurrence (95% CI)	P-value	Odds ratio for cluster occurrence (95% CI)	P-value cluster
Midazolam	1.28 (0.93–1.75)	0.13	1.35 (1–1.81)	0.048
Fentanyl	0.97 (0.68–1.38)	0.86		
Propofol	0.89 (0.68–1.16)	0.38	0.68 (0.49–0.95)	0.03
Sufentanil	1.06 (0.64–1.76)	0.83		
Ketamine	0.38 (0.18–0.79)	0.01	0.2 (0.06–0.64)	0.01
Morphine	0.93 (0.65–1.3)	0.67		

Drugs were included in the model using the enter method. Relative risk for the occurrence of spreading depolarizations and spreading depolarization clusters are shown for dose ranges of each drug in quartiles. For cluster analysis, the table includes stepwise backward selected drugs.

spreading depolarization occurrence during hours recorded with ketamine was not associated with any of the total recorded hours, the total recorded hours with spreading depolarizations or the time after ictus (Fig. 2B). To further evaluate a dose–response relationship, all given doses of ketamine were correlated with their spreading depolarization frequencies. A significant linear association was found between ketamine dose and spreading depolarization frequency (Fig. 2C). On the basis of the fact that this study was fully observational, and none of the drugs were given with the intention to treat spreading depolarizations, our preliminary outcome analysis (data not shown) must be viewed with caution. Patients with high spreading depolarization per hour frequencies displayed a trend towards bad outcome (Spearman $r = -0.2$; $P < 0.1$, $n = 105$ patients); however, although ketamine reduces spreading depolarization per hour frequencies (Fig. 2C), increasing doses of ketamine were correlated with poorer extended Glasgow Outcome Scale after 6 months (Spearman $r = -0.4$; $P < 0.001$, $n = 105$ patients).

The best logistic regression model to predict the influence of analgesic and sedative drugs on clusters of spreading depolarizations was chosen by the backward stepwise elimination technique. This analysis revealed a significant positive association between midazolam administration and cluster occurrence and a significant negative association between ketamine administration and cluster occurrence (Table 4). Propofol was also negatively associated with cluster occurrence, although not to the same extent as ketamine (Table 4).

Isoelectric spreading depolarizations are believed to represent a subtype of spreading depolarizations that occur in either ischaemic or immediately peri-ischaemic tissue, the ischaemic part of which being at risk for cell death with permanent loss of neuronal function (Oliveira-Ferreira et al., 2010). Of 1340 total hours of recording, 127 h with isoelectric spreading depolarization events in 13 patients were observed. Unfortunately, these data are too limited to allow the use of a multivariate statistical model for analysis. However, calculations of univariate, patient-independent odds ratios revealed a significant decrease in the probability of isoelectric spreading depolarizations for most drugs (Fig. 2D).

Because the injured brain has general mechanisms of reaction and repair such as oedema, swelling and neuronal reorganization, it seemed reasonable to evaluate the overall occurrence of spreading depolarizations following different kinds of brain insult, such as traumatic brain injury, malignant hemispheric stroke, subarachnoid

haemorrhage and intracerebral haemorrhage. However, it remains possible that the observed effect of ketamine and other drugs on spreading depolarizations could be limited to one or a few specific types of brain injury. Indeed, we found that spreading depolarization occurrence within the different disease entities showed different responses to drug treatment. Still, the relatively small number of patients per disease group limited the power of our analysis. This was reflected by the large variability within the recorded data. Within recordings from patients with traumatic brain injury, we found a peak of recorded hours with and without drug at Day 2 after ictus. The recordings from patients with subarachnoid haemorrhage and intracerebral haemorrhage peaked on Day 4 after ictus, and most recordings after malignant hemispheric stroke were made on Day 5 after ictus (Fig. 3A). The recording time from patients with malignant hemispheric stroke without influence of one of the investigated drugs was low. Only a total of 268 recorded hours between Days 1 and 8 were obtained without analgesia or sedation (Fig. 3B), and none of these contained spreading depolarizations. On the basis of the number of spreading depolarizations observed following malignant hemispheric stroke under the influence of analgesics and sedatives, we would have expected to observe at least one spreading depolarization in 11 h of recordings without drugs. Nonetheless, we attribute this surprising observation to be an artefactual consequence of the small number of recorded hours without drug influence. In the subarachnoid haemorrhage/intracerebral haemorrhage and traumatic brain injury disease groups, we found that for most days after ictus, the number of spreading depolarizations/hour recorded under analgesics and sedatives are well below that observed for recordings made without sedation (Fig. 3B). The effect of ketamine on spreading depolarization occurrence was sustained for all disease groups (Fig. 3C). Other drugs also seemed to influence spreading depolarization occurrence in this subgroup analysis. Because these results are based on relatively low patient numbers, however, their value is limited.

Discussion

The most conspicuous finding of this analysis was a strong and sustained suppression of spreading depolarizations by ketamine. Specifically, administration of ketamine was found to be inversely correlated with the occurrence of spreading depolarizations,
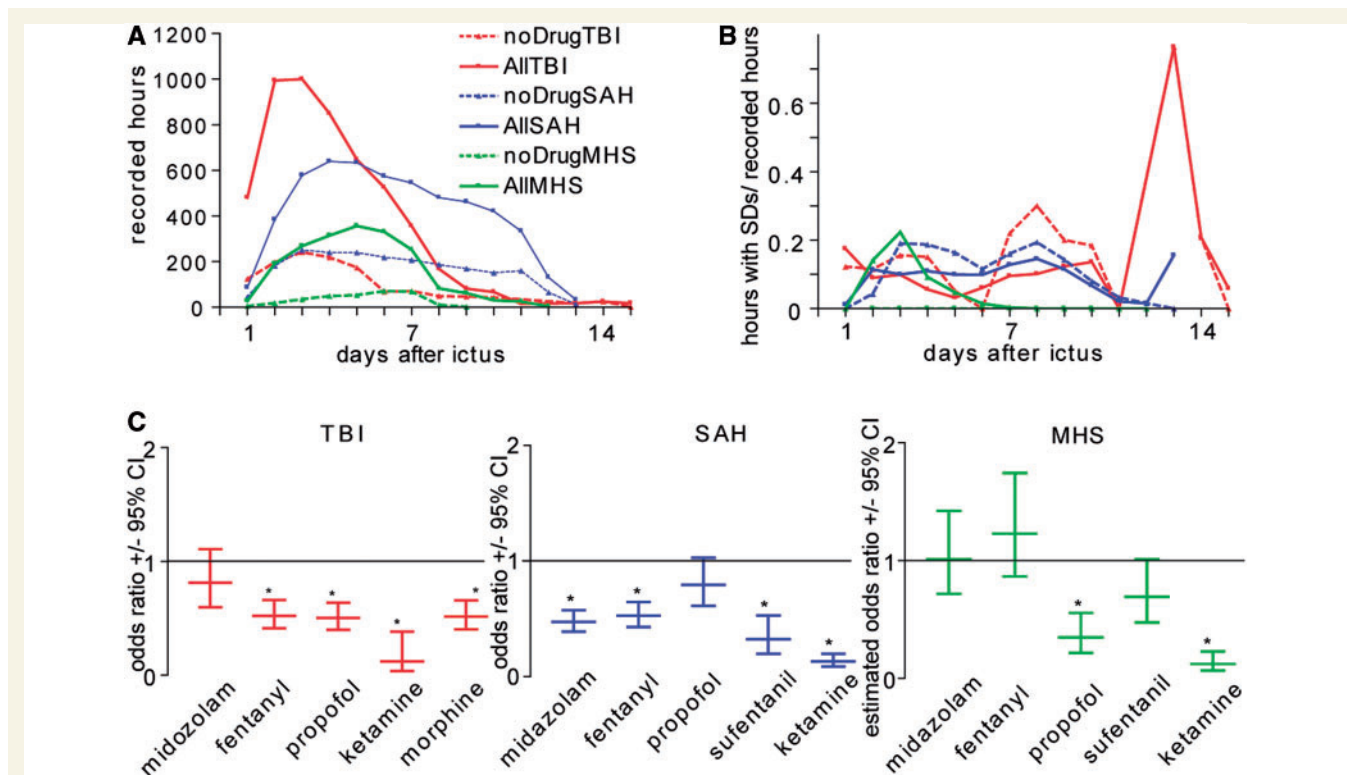
Figure 3 Analgesics and sedatives influence the relative probability of spreading depolarization (SDs) occurrence after traumatic brain injury (TBI), subarachnoid haemorrhage (SAH) and malignant hemispheric stroke (MHS). (**A**) The hours of electrocorticographic recordings obtained after acute brain injury were highly variable and depended on the type of brain injury. (**B**) Rates of spreading depolarization occurrence for all recorded hours and for all hours recorded without the investigated drugs. (**C**) Analgesic and sedative drugs changed the relative probability of spreading depolarization occurrence in a disease- and drug-specific manner. *Significant at $P < 0.05$. noDrug = none of the drugs were used; All = all recording hours are included.

clusters of spreading depolarizations, and isoelectric spreading depolarizations. This effect was observed for all disease-specific subgroups and was persistent across all days after the initial event of brain injury.

To date only two patients with traumatic brain injury and aneurysmal subarachnoid haemorrhage were reported in whom spreading depolarizations were abolished under the influence of the non-competitive NMDA receptor antagonist ketamine (Sakowitz et al., 2009). Another patient with aneurysmal subarachnoid haemorrhage displayed a cluster of spreading depolarizations under a lower dose of ketamine, developed severe delayed ischaemic strokes and died (Dreier et al., 2009). These heterogeneous findings may reflect a situation, recognized from animal experiments, that deleterious conditions induce spreading depolarization as they shift the balance of ion fluxes across neuron cell membranes towards a net positive inward flux, whereas protective drugs such as NMDA receptor antagonists have the opposite effect. If the deleterious condition is stronger than the NMDA receptor antagonist, spreading depolarization will occur despite the presence of the drug in the region exposed to the deleterious condition (Hernándéz-Cáceres et al., 1987; Aitken et al., 1988); nevertheless, the NMDA receptor antagonist may be sufficient to block the spread of spreading depolarization into the healthier peri-ischaemic tissue. Spreading

depolarizations may have beneficial effects in the peri-ischaemic tissue where they could have preconditioning effects or promote plasticity and regeneration (Nakamura et al., 2010; Dreier, 2011). Theoretically, this suggests the risk that spreading depolarizations will be unaffected by an underdosed NMDA receptor antagonist in regions where they lead to damage but will be abolished where they could be beneficial. However, in this study, we found a strong and sustained effect on spreading depolarizations. In addition, an effect on isoelectric spreading depolarizations was observed. Isoelectric spreading depolarizations occur immediately not only in the peri-ischaemic tissue but also in the ischaemic zone where spreading depolarizations are deleterious (Oliveira-Ferreira et al., 2010; Hartings et al., 2011a). Consistent with the notion that isoelectric spreading depolarizations carry a particular risk, it has been recently found that they were associated with a worse outcome after traumatic brain injury (Hartings et al., 2011b). So far, clinical evidence regarding neurological recovery after ketamine administration is limited. Ketamine has been shown to increase cerebral perfusion pressure and might therefore increase neuronal survival (Kolenda et al., 1996; Albanese et al., 1997). However, an improved outcome has not been reported (Roberts et al., 2011). Underdosing of the drug and adverse effects were possibly responsible for the failure of NMDA receptor antagonists in a number of clinical studies on

stroke and brain trauma (Lees et al., 2000; Sacco et al., 2001; Ikonomidou and Turski, 2002). Despite this failure, ketamine is still associated with a powerful neuroprotective potential (Hudetz and Pagel, 2010). Neuromonitoring of spreading depolarizations may offer the chance for real-time detection of the advent of ischaemic complications in patients with subarachnoid haemorrhage or traumatic brain injury. This could allow early targeted treatment for those patients who benefit from such therapy, whereas drug treatment with unnecessary side effects is avoided in those patients who do not develop the complication (Hartings et al., 2012). Thus, it may be worthwhile to retest the potential neuroprotective effect of an NMDA receptor antagonist such as ketamine in patients in whom depolarizations are detected.

On a cellular level, spreading depolarizations reflect a breakdown of ion homeostasis in neurons and glia (Hopwood et al., 2005). Although functioning and viable glia are believed to be beneficial for neuronal survival, they seem not to be critical for the generation of spreading depolarizations (Largo et al., 1996). Spreading depolarization may result either from acute hyperexcitability with increased influx of cations, or from disturbance of $Na+/K+$-ATPase activity with decreased outflux of cations. Thus, spreading depolarization starts when net cation fluxes of sodium and calcium turn inwards, being no longer compensated by the outward flux produced by $Na+/K+$-ATPase activity (Kager et al., 2002). Ischaemia is an important mechanism underlying reduced $Na+/K+$-ATPase activity as ATP is no longer able to drive the pump. Cation influx across the neuronal membrane is triggered by neuronal activity and by second messengers and ligands. Anaesthetics and sedative drugs primarily target receptors on neuronal cells that influence ion homeostasis and neuronal excitability. Ketamine, for instance, binds to the NMDA receptor, and during glutamatergic synaptic transmission, it decreases its open probability—and therewith the total sodium influx into a cell. Indeed, the use of non-competitive NMDA receptor antagonists is considered one of the most promising strategies for suppression of spreading depolarizations (Iijima et al., 1992; Obrenovitch and Zilkha, 1996). The first experimental evidence to this effect was obtained in animal experiments and was attributed specifically to NMDA receptor blockade (Marrannes et al., 1988). Human brain slices obtained after neurosurgical resection for epilepsy likewise showed diminished spreading depolarizations after treatment with antagonists of the NMDA receptor (Avoli et al., 1991; Gorji et al., 2001).

Our analyses suggest that other commonly used intensive care unit drugs exert little to no influence on the occurrence of spreading depolarizations. Besides ketamine, midazolam displayed a possible influence on spreading depolarizations and was associated with increased numbers of spreading depolarization clusters. Midazolam, like all benzodiazepines, enhances GABAergic activity and leads to neuronal hyperpolarization. This reduction in neuronal excitability should theoretically lead to less spreading depolarizations and cannot be responsible for increased clusters of spreading depolarizations. But there are other possible explanations for this observation. Midazolam and ketamine have opposite effects on brain energy consumption, and brain metabolism plays a crucial role in generation and propagation of spreading depolarizations. Specifically, the reduction of cerebral metabolic rate under the influence of midazolam is magnitudes greater than that measured during normal brain inactivity without the influence of sedatives (Alkire et al., 1999; Shulman et al., 2009). Ketamine has been shown to increase energy consumption of the brain (Dawson et al., 1971; Långsjö et al., 2003). In contrast with the effect of ketamine on brain metabolism and spreading depolarization clusters, midazolam might induce clusters of spreading depolarizations along with the reduction of brain energy utilization. These findings suggest that GABAergically mediated metabolic depression could potentially aggravate the failure of energy-dependent ion pumps and therefore increase generation and propagation of spreading depolarization clusters.

The analysis of spreading depolarization frequency (spreading depolarization per hour) with and without each drug was used as a straightforward approach. Using continuous spreading depolarization per hour data, we analysed spreading depolarization occurrence with and without anaesthetic or sedative drugs (Fig. 2A). With respect to drug dosage, we analysed hours with spreading depolarizations including hours with one or more spreading depolarization. To resolve the important occurrence of multiple spreading depolarizations within close proximity, clusters of three or more spreading depolarizations were further analysed (Feuerstein et al., 2010). Patients in this retrospective study received no standard dose or standard drug regimen. Thus, changes in drug dosage and in sedative or opioid were common during recordings, even in a single patient. In addition, spreading depolarizations are rare events and do not even occur in all patients. Taken together, the variability of the available data and the number of patients included limited the power of our analysis. Furthermore, neuroprotective properties of ketamine could not be characterized in our patients. A part of this may be due to the fact that sedative use per se is correlated with the extent of the initial brain injury, i.e. patients with severe brain tissue injury are more likely to require sedation during neurointensive care treatment. We were nonetheless surprised by this result given that ketamine reduced spreading depolarization frequency, and a reduced spreading depolarization frequency was generally associated with a better outcome.

In this observational study, we proposed that a cluster of spreading depolarizations should be defined as the occurrence of three or more spreading depolarizations within 3 h. It is reasonable to distinguish isolated spreading depolarizations from temporally clustered spreading depolarizations as the latter are likely to be more harmful to brain tissue (Dreier et al., 2006; Bosche et al., 2010). Future investigations may provide a better indication of what number and frequency of clustered spreading depolarizations compromise metabolism critically and result in irreversible damage to functional brain tissue. Data presented here suggest, however, that a cluster must consist of at least three spreading depolarizations within close temporal proximity, as the observation of any fewer or more widely distributed spreading depolarizations in a cluster may be merely coincidental.

Conclusion

Analgesic and sedative drugs had a significant impact on the occurrence of spreading depolarizations. In particular, ketamine

markedly decreased the probability for cortical spreading depolar-izations to occur. This finding suggests that spreading depolariza-tions can be modulated in humans and are therefore valuable targets for neuroprotective therapy that might even exceed the action of ketamine. Prospective clinical studies with neuromonitor-ing of spreading depolarizations are needed to verify both the effects of ketamine on spreading depolarization occurrence and to examine its benefits for acutely brain-injured patients.

Acknowledgements

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References

Aitken PG, Balestrino M, Somjen GG. NMDA antagonists: lack of protective effect against hypoxic damage in CA1 region of hippocam-pal slices. Neurosci Lett 1988; 89: 187–92.

Albanèse J, Arnaud S, Rey M, Thomachot L, Alliez B, Martin C. Ketamine decreases intracranial pressure and electroencephalographic activity in traumatic brain injury patients during propofol sedation. Anesthesiology 1997; 87: 1328–34.

Alkire MT, Pomfrett CJ, Haier RJ, Gianzero MV, Chan CM, Jacobsen BP, et al. Functional brain imaging during anesthesia in humans: effects of halothane on global and regional cerebral glucose metabolism. Anesthesiology 1999; 90: 701–9.

Avoli M, Drapeau C, Louvel J, Pumain R, Olivier A, Villemure JG. Epileptiform activity induced by low extracellular magnesium in the human cortex maintained in vitro. Ann Neurol 1991; 30: 589–96.

Bosche B, Graf R, Ernestus RI, Dohmen C, Reithmeier T, Brinker G, et al. Members of the Cooperative Study of Brain Injury Depolarizations (COSBID). Recurrent spreading depolarizations after subarachnoid hemorrhage decreases oxygen availability in human cerebral cortex. Ann Neurol 2010; 67: 607–17.

Canals S, Makarova I, López-Aguado L, Largo C, Ibarz JM, Herreras O. Longitudinal depolarization gradients along the somatodendritic axis of CA1 pyramidal cells: a novel feature of spreading depression. J Neurophysiol 2005; 94: 943–51.

Dawson B, Michenfelder JD, Theye RA. Effects of ketamine on canine cerebral blood flow and metabolism: modification by prior administra-tion of thiopental. Anesth Analg 1971; 50: 443–7.

Dohmen C, Sakowitz OW, Fabricius M, Bosche B, Reithmeier T, Ernestus RI, et al. Co-Operative Study of Brain Injury Depolarisations (COSBID). Spreading depolarizations occur in human ischemic stroke with high incidence. Ann Neurol 2008; 63: 720–8.

Dreier JP. The role of spreading depression, spreading depolarization and spreading ischemia in neurological disease. Nat Med 2011; 17: 439–47.

Dreier JP, Major S, Manning A, Woitzik J, Drenckhahn C, Steinbrink J, et al. COSBID study group. Cortical spreading ischaemia is a novel process involved in ischaemic damage in patients with aneurysmal subarachnoid haemorrhage. Brain 2009; 132 (Pt 7): 1866–81.

Dreier JP, Woitzik J, Fabricius M, Bhatia R, Major S, Drenckhahn C, et al. Delayed ischaemic neurological deficits after subarachnoid haemor-rhage are associated with clusters of spreading depolarizations. Brain 2006; 129 (Pt 12): 3224–37.

Fabricius M, Fuhr S, Bhatia R, Boutelle M, Hashemi P, Strong AJ, et al. Cortical spreading depression and peri-infarct depolarization in acutely injured human cerebral cortex. Brain 2006; 129 (Pt 3): 778–90.

Feuerstein D, Manning A, Hashemi P, Bhatia R, Fabricius M, Tolias C, et al. Dynamic metabolic response to multiple spreading depolariza-tions in patients with acute brain injury: an online microdialysis study. J Cereb Blood Flow Metab 2010; 30: 1343–55.

Gorji A, Scheller D, Straub H, Tegtmeier F, Köhling R, Höhling JM, et al. Spreading depression in human neocortical slices. Brain Res 2001; 906: 74–83.

Hartings JA, Bullock MR, Okonkwo DO, Murray L, Murray GD, Fabricius M, et al. Cortical spreading depolarizations are a novel mechanism independently associated with unfavorable neurologic outcome from traumatic brain injury. Lancet Neurol 2011b; 10: 1058–64.

Hartings JA, Strong AJ, Okonkwo DO, Bullock MR. Spreading depolar-isations and traumatic brain injury: time course and mechanisms. Lancet Neurol 2012; 11: 389–90.

Hartings JA, Watanabe T, Bullock MR, Okonkwo DO, Fabricius M, Woitzik J, et al. for the Co-Operative Study on Brain Injury Depolarizations. Spreading depolarizations have prolonged direct cur-rent shifts and are associated with poor outcome in brain trauma. Brain 2011a; 134 (Pt 5): 1529–40.

Hernández-Cáceres J, Macias-González R, Brozek G, Bures J. Systemic ketamine blocks cortical spreading depression but does not delay the onset of terminal anoxic depolarization in rats. Brain Res 1987; 437: 360–4.

Hopwood SE, Parkin MC, Bezzina EL, Boutelle MG, Strong AJ. Transient changes in cortical glucose and lactate levels associated with peri-infarct depolarisations, studied with rapid-sampling microdialysis. J Cereb Blood Flow Metab 2005; 25: 391–401.

Hudetz JA, Pagel PS. Neuroprotection by ketamine: a review of the experimental and clinical evidence. J Cardiothorac Vasc Anesth 2010; 24: 131–42.

Iijima T, Mies G, Hossmann KA. Repeated negative DC deflections in rat cortex following middle cerebral artery occlusion are abolished by MK-801: effect on volume of ischemic injury. J Cereb Blood Flow Metab 1992; 12: 727–33.

Ikonomidou C, Turski L. Why did NMDA receptor antagonists fail clinical trials for stroke and traumatic brain injury? Lancet Neurol 2002; 1: 383–6.

Kager H, Wadman WJ, Somjen GG. Conditions for the triggering of spreading depression studied with computer simulations. J Neurophysiol 2002; 88: 2700–12.

Kolenda H, Gremmelt A, Rading S, Braun U, Markakis E. Ketamine for analgosedative therapy in intensive care treatment of head-injured patients. Acta Neurochir (Wien) 1996; 138: 1193–9.

Långsjö JW, Kaisti KK, Aalto S, Hinkka S, Aantaa R, Oikonen V, et al. Effects of subanesthetic doses of ketamine on regional cerebral blood flow, oxygen consumption, and blood volume in humans. Anesthesiology 2003; 99: 614–23.

Largo C, Cuevas P, Somjen GG, Martín del Río R, Herreras O. The effect of depressing glial function in rat brain in situ on ion homeostasis, synaptic transmission, and neuron survival. J Neurosci 1996; 16: 1219–29.

Lauritzen M, Dreier JP, Fabricius M, Hartings JA, Graf R, Strong AJ. Clinical relevance of cortical spreading depression in neurological disorders: migraine, malignant stroke, subarachnoid and intracranial hemorrhage, and traumatic brain injury. J Cereb Blood Flow Metab 2011; 31: 17–35.

Lees KR, Asplund K, Carolei A, Davis SM, Diener HC, Kaste M, et al. Glycine antagonist (gavestinel) in neuroprotection (GAIN International) in patients with acute stroke: a randomised controlled trial. GAIN International Investigators. Lancet 2000; 355: 1949–54.

Marrannes R, Willems R, De Prins E, Wauquier A. Evidence for a role of the N-methyl-D-aspartate (NMDA) receptor in cortical spreading depression in the rat. Brain Res 1988; 457: 226–40.

Nakamura H, Strong AJ, Dohmen C, Sakowitz OW, Vollmar S, Sué M, et al. Spreading depolarizations cycle around and enlarge focal ischae-mic brain lesions. Brain 2010; 133 (Pt 7): 1994–2006.

Obrenovitch TP, Zilkha E. Inhibition of cortical spreading depression by L-701,324, a novel antagonist at the glycine site of the N-methyl-D-aspartate receptor complex. Br J Pharmacol 1996; 117: 931–7.

Oliveira-Ferreira AI, Milakara D, Alam M, Jorks D, Major S, Hartings JA, et al. COSBID study group. Experimental and preliminary clinical evidence of an ischemic zone with prolonged negative DC shifts sur-rounded by a normally perfused tissue belt with persistent electrocor-ticographic depression. J Cereb Blood Flow Metab 2010; 30: 1504–19.

Roberts DJ, Hall RI, Kramer AH, Robertson HL, Gallagher CN, Zygun DA. Sedation for critically ill adults with severe traumatic brain injury: a systematic review of randomized controlled trials. Crit Care Med 2011; 39: 2743–51.

Sacco RL, DeRosa JT, Haley EC Jr, Levin B, Ordronneau P, Phillips SJ, et al. Glycine Antagonist in Neuroprotection Americas Investigators. Glycine antagonist in neuroprotection for patients with acute stroke: GAIN Americas: a randomized controlled trial. JAMA 2001; 285: 1719–28.

Sakowitz OW, Kiening KL, Krajewski KL, Sarrafzadeh AS, Fabricius M, Strong AJ, et al. Preliminary evidence that ketamine inhibits spreading depolarizations in acute human brain injury. Stroke 2009; 40: e519–e522.

Shulman RG, Hyder F, Rothman DL. Baseline brain energy supports the state of consciousness. Proc Natl Acad Sci USA 2009; 106: 11096–101.

Somjen GG. Mechanisms of spreading depression and hypoxic spreading depression-like depolarization. Physiol Rev 2001; 81: 1065–96. Review.

Strong AJ, Fabricius M, Boutelle MG, Hibbins SJ, Hopwood SE, Jones R, et al. Spreading and synchronous depressions of cortical activity in acutely injured human brain. Stroke 2002; 33: 2738–43.

BRAIN
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Correlates of spreading depolarization in human scalp electroencephalography

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It has been known for decades that suppression of spontaneous scalp electroencephalographic activity occurs during ischaemia. Trend analysis for such suppression was found useful for intraoperative monitoring during carotid endarterectomy, or as a screening tool to detect delayed cerebral ischaemia after aneurismal subarachnoid haemorrhage. Nevertheless, pathogenesis of such suppression of activity has remained unclear. In five patients with aneurismal subarachnoid haemorrhage and four patients with decompressive hemicraniectomy after malignant hemispheric stroke due to middle cerebral artery occlusion, we here performed simultaneously full-band direct and alternating current electroencephalography at the scalp and direct and alternating current electrocorticography at the cortical surface. After subarachnoid haemorrhage, 275 slow potential changes, identifying spreading depolarizations, were recorded electrocorticographically over 694 h. Visual inspection of time-compressed scalp electroencephalography identified 193 (70.2%) slow potential changes [amplitude: -272 (-174, -375) μV (median quartiles), duration: 5.4 (4.0, 7.1) min, electrocorticography–electroencephalography delay: 1.8 (0.8, 3.5) min]. Intervals between successive spreading depolarizations were significantly shorter for depolarizations with electroencephalographically identified slow potential change [33.0 (27.0, 76.5) versus 53.0 (28.0, 130.5) min, $P = 0.009$]. Electroencephalography was thus more likely to display slow potential changes of clustered than isolated spreading depolarizations. In contrast to electrocorticography, no spread of electroencephalographic slow potential changes was seen, presumably due to superposition of volume-conducted electroencephalographic signals from widespread cortical generators. In two of five patients with subarachnoid haemorrhage, serial magnetic resonance imaging revealed large delayed infarcts at the recording site, while electrocorticography showed clusters of spreading depolarizations with persistent depression of spontaneous activity. Alternating current electroencephalography similarly displayed persistent depression of spontaneous activity, and direct current electroencephalography slow potential changes riding on a shallow negative ultraslow potential. Isolated spreading depolarizations with depression of both spontaneous electrocorticographic and electroencephalographic activity displayed significantly longer intervals between successive spreading depolarizations than isolated depolarizations with only depression of electrocorticographic activity [44.0 (28.0,

132.0) min, *n* = 96, versus 30.0 (26.5, 51.5) min, *n* = 109, *P* = 0.001]. This suggests fusion of electroencephalographic depression periods at high depolarization frequency. No propagation of electroencephalographic depression was seen between scalp electrodes. Durations/magnitudes of isolated electroencephalographic and corresponding electrocorticographic depression periods correlated significantly. Fewer spreading depolarizations were recorded in patients with malignant hemispheric stroke but characteristics were similar to those after subarachnoid haemorrhage. In conclusion, spreading depolarizations and depressions of spontaneous activity display correlates in time-compressed human scalp direct and alternating current electroencephalography that may serve for their non-invasive detection.

Introduction

In the last decades, hundreds of Phase II and III clinical trials on presumed neuroprotective agents for stroke have failed (see Washington University Internet Stroke Centre, www.strokecenter.org). Therefore, a new road map for neuroprotection has been proposed including better proof of efficacy of neuroprotectants in animal models and efficacy in novel clinical proof of concept studies (Donnan, 2008). In animal experiments, neuroprotectants have been found to be most effective either when given within minutes after the onset of acute neuronal injury or when they have been pre-administered. Moreover, recent experimental evidence suggests that phases of acute injury and repair may require opposing neuroprotective strategies (Lo, 2008). It would thus be promising to develop clinical proof of concept studies where a biomarker can distinguish between phases of acute injury and repair to guide phase-specific treatment allocation of neuroprotectants. Moreover, the biomarker ought to allow a read-out of the neuroprotectant's effect on the parenchyma. Ideally, such a biomarker should reflect neuronal injury in real-time and should be measurable non-invasively.

Spreading depolarization is the mechanism of the abruptly developing cytotoxic oedema in cerebral grey matter (Klatzo, 1987; Dreier, 2011). Spreading depolarization occurs when neuronal cation efflux by the ATP-dependent sodium pumps locally fails to compensate for cation influx of sodium and calcium (Somjen, 2001). The ion fluxes result in a net gain of solutes by the neurons accompanied by water influx. Specifically, spreading depolarization is characterized as a wave of massive ion translocation between intra- and extracellular space, near-complete sustained neuronal depolarization, glial depolarization and neuron swelling. Spreading depolarization is observed as an abrupt, large, negative slow potential change as measured in the low-frequency or direct current (DC) range of the electrocorticogram (Canals *et al.*, 2005; Oliveira-Ferreira *et al.*, 2010; Hartings *et al.*, 2011*b*). In the high-frequency or alternating current (AC) range of the electrocorticography, spreading depolarization causes silencing of spontaneous activity (spreading depression).

Spreading depolarization is induced by energy depletion as well as other stimuli such as chemicals, neurotransmitters and mechanical damage. Spreading depolarization may be followed by either recovery, dependent on sufficient recruitment of sodium pump

activity (LaManna and Rosenthal, 1975), or neuronal death (Takano *et al.*, 2007; Risher *et al.*, 2010; Dreier, 2011). Progressive injury and cell death are manifested in a negative ultraslow DC potential, which is possibly of glial and neuronal origin (Herreras and Somjen, 1993; Somjen, 2001; Oliveira-Ferreira *et al.*, 2010). Hence, real-time detection of infarction can be achieved using electrodes on the brain surface over the newly developing infarct through measurement of the large negative ultraslow DC potential on which one or more slow potential changes/spreading depolarizations are riding (Oliveira-Ferreira *et al.*, 2010). However, spreading depolarizations typically propagate away from or circle around the infarcted tissue at a speed of ~3 mm/min (Nakamura *et al.*, 2010). Even remote from the actual infarct, recording electrodes may therefore identify occurrence of a new infarct when a cluster of recurrent, short-lasting spreading depolarizations is recorded (Dreier *et al.*, 2006; Dohmen *et al.*, 2008). Spreading depolarizations of such a cluster ride on a shallow negative ultraslow potential in the ischaemic penumbra (current sink), whereas they ride on a shallow positive potential in the normally perfused surrounding tissue (current source) (Oliveira-Ferreira *et al.*, 2010). Between recurrent spreading depolarizations, normally perfused tissue immediately around the penumbra shows persistent depression of spontaneous activity in similar fashion to the penumbra, whereas repeated cycles of spreading depression of spontaneous activity followed by recovery are observed further away (Oliveira-Ferreira *et al.*, 2010; Hartings *et al.*, 2011*b*). Whether and how zonal gradients of the slow potentials along laminar profiles across the cortex reflect such interregional electrophysiological differences in and around ischaemic zones has not been studied sufficiently while current sources and sinks are well established for epileptic seizure activity and spreading depolarization in otherwise healthy tissue (Wadman *et al.*, 1992).

Currently, clinical monitoring of spreading depolarizations is limited to patients who require neurosurgical interventions that allow for placement of a subdural electrode strip such as surgical aneurysm ligation after subarachnoid haemorrhage (SAH), placement of extraventricular drainage, decompressive hemicraniectomy or evacuation of a haematoma (Strong *et al.*, 2002; Dreier *et al.*, 2006; Dohmen *et al.*, 2008). Thus, spreading depolarizations have been recorded in abundance in individuals with aneurismal SAH, delayed ischaemic stroke after aneurismal SAH, malignant

hemispheric stroke (MHS), spontaneous intracerebral haemorrhage or traumatic brain injury (Strong *et al.*, 2002; Dreier *et al.*, 2006, 2009; Fabricius *et al.*, 2006; Dohmen *et al.*, 2008; Hartings *et al.*, 2011*b*). Nevertheless, the monitoring of spreading depolarizations could be extended to other patient populations if non-invasive technology allowed their reliable detection. Here, we investigated whether ultraslow DC potential, slow potential change and/or depression of spontaneous activity can be recorded using scalp DC/AC-EEG while occurrence of those signals was validated at the brain surface using invasive near-DC/AC-electrocorticography or DC/AC-electrocorticography.

Patients and methods

Patient recruitment

Five patients with aneurismal SAH and four patients with MHS were recruited at two centres of the Charité University Medicine Berlin participating in the Co-Operative Studies on Brain Injury Depolarizations (COSBID) [Campus Virchow Klinikum ($n = 5$) and Campus Benjamin Franklin ($n = 4$)]. Inclusion criteria were age $\geqslant 18$ years and the clinical decision for either craniotomy to perform surgical ligation of an aneurysm, evacuation of a haematoma and/or decompression, or for an extended burr hole to place a ventricular drain or oxygen sensor. Patients with fixed, dilated pupils were excluded. Research protocols were approved by the institutional review board and surrogate informed consent was obtained for all patients. All research was conducted in accordance with the Declaration of Helsinki. Aneurismal SAH and MHS were diagnosed by the assessment of CT and CT angiography scans. In three patients with aneurismal SAH, additional digital subtraction angiography was performed initially.

At the conclusion of surgery, a single, linear, six-contact (platinum) recording strip (Wyler, 5 mm diameter; Ad-Tech Medical) was placed on the surface of the cortex for subsequent electrocorticography recordings (Strong *et al.*, 2002; Dreier *et al.*, 2006; Dohmen *et al.*, 2008). After surgery, patients were transferred to the intensive care unit where continuous electrocorticography and EEG recordings were acquired for up to 15 days after aneurismal SAH and up to 8 days after MHS. Thereafter, electrode strips were removed at the bedside by gentle traction. No haemorrhagic or infectious complications of the electrode strip were encountered. Clinical outcome was assessed using the modified Rankin Scale on Day 15 after aneurismal SAH and on Day 8 after MHS.

Throughout recordings, patients were ventilated and pharmacologically immobilized as required. Sedation was mostly maintained with propofol or midazolam, and analgesia was provided with fentanyl. Intracranial pressure was monitored, if clinically indicated, by a ventricular drainage catheter or an intraparenchymal intracranial pressure transducer (Codman). Glasgow Coma Score, blood gases, glucose and electrolytes were documented at least every 6 h. A thorough neurological examination was performed at least daily.

Patients with aneurismal subarachnoid haemorrhage

Clinical presentation of aneurismal SAH was classified according to the World Federation of Neurological Surgeons scale. Amount and distribution of subarachnoid blood was graded according to the Fisher scale (Kistler *et al.*, 1983). After diagnosis of aneurismal SAH,

surgical and/or endovascular interventions were performed within the next 24 h. Three patients underwent surgical clipping, one endovascular coiling and one was initially coiled followed by clipping due to aneurysm rupture in the course of coiling. The recording strip was positioned on viable, but often oedematous tissue.

A delayed ischaemic neurological deficit was defined as the occurrence of focal neurological impairment (such as hemiparesis, aphasia, apraxia, hemianopia or neglect), or a decrease of at least two Glasgow Coma Scale points [either on the total score or on one of its individual components (eye, motor on either side, verbal)]. Moreover, the diagnosis of a delayed ischaemic neurological deficit required that the neurological deficit was not present immediately after aneurysm occlusion, that it lasted for at least 1 h, could not be attributed to other causes such as hydrocephalus or re-bleeding by means of clinical assessment, CT or MRI of the brain, and appropriate laboratory studies, and did not occur earlier than 72 h after the initial haemorrhage (Vergouwen *et al.*, 2010). Serial MRI scans were performed postoperatively, at the time of clinical deterioration and after the monitoring period to screen for delayed infarcts. Admission and follow-up neuroimages were independently evaluated by a study neuroradiologist (M.S.), blinded to the electrocorticography data, for the presence of focal or global cerebral oedema, the Fisher grade, the presence and degree of hydrocephalus, the presence of infarction, intracerebral or subdural haematoma.

Criteria for proximal vasospasm after aneurismal SAH were defined using digital subtraction angiography as $>30\%$ narrowing of the arterial luminal diameter in one of the following arterial segments: A1, A2, M1, M2 and C1–C2. Magnification errors were corrected by comparing extradural segments of the internal carotid artery (C4–C5). Using transcranial Doppler sonography, significant vasospasm was defined by a mean velocity $\geqslant 200$ cm/s in at least one middle cerebral artery (Vora *et al.*, 1999). Vasospasm was excluded if the middle cerebral artery mean velocities remained <120 cm/s throughout the observation period.

Patients with delayed cerebral ischaemia were treated with haemodynamic therapy (hypertension, hypervolaemia) (van Gijn and Rinkel, 2001). Oral nimodipine was given prophylactically in all patients.

Patients with malignant hemispheric stroke

Clinical presentation of MHS was classified according to the National Institute of Health Stroke Scale. All patients with MHS suffered from $\geqslant 2/3$ infarction of the middle cerebral artery territory. Hemicraniectomy was initiated following similar in- and exclusion criteria as recently published for DESTINY II (Juttler *et al.*, 2011), but inclusion criterion for age was $\geqslant 18$ years. In patients with MHS, the electrode strip was implanted over presumed viable peri-infarct tissue of the anterior cerebral artery. The electrode was advanced under the bone rim to ensure proper fixation so that the strip was placed tangentially in relation to the border of infarction. The lead wire of the strip was externalized through a burr hole in the skull (if the bone flap was replaced) and tunnelled beneath the scalp to exit 2–3 cm from the craniotomy margin.

Near-DC/AC- and DC/AC-electrocorticography recordings at the cortical surface

Two ipsilateral, subdermal platinum needle electrodes (SpesMedica) served as reference and ground for the invasive, subdural recordings.

Near-DC/AC-electrocorticography signals were recorded by a GT205 amplifier (bandpass: 0.01–45 Hz, sampling rate: 200 Hz) connected to a Powerlab 16/SP analogue/digital converter (ADInstruments). In some patients, electrocorticography was measured in parallel using a BrainAmp amplifier in order to record DC in addition to near-DC potential components (bandpass: 0–1000 Hz, sampling rate: 2500 Hz, BrainAmp MR plus, Brain Products). Subdural electrodes were connected in sequential bipolar fashion as well as in unipolar fashion, each referenced to the ipsilateral subdermal platinum electrode. Data were recorded and reviewed with the use of LabChart 7 software (ADInstruments) and BrainVision Recorder 1.05 software (Brain Products), respectively.

Scalp DC/AC-EEG recordings

In order to monitor the full-band DC/AC–EEG (bandpass: 0–1000 Hz, sampling rate: 2500 Hz), sintered Ag/AgCl electrodes (EasyCap) were connected to a BrainAmp amplifier. The number of recording electrodes varied from 8 to 13 depending on the scalp area shaved for the neurosurgical procedure and the localization of exit points from the skull for invasive probes such as the external ventricular drain. Ipsilateral to the electrocorticography strip, 6–8 scalp electrodes were placed covering the frontal, parietal and temporal brain regions; 2–5 electrodes were placed contralaterally. Electrodes were positioned in accordance to the international 10–20 electrode system (Klem *et al.*, 1999). The reference electrode was placed on the mastoid ipsilateral to the electrocorticography recording strip. An electrode in the frontal midline served as ground. The electrodes were attached with Collodion adhesive (Mavidon). Abrasive electrode gel (Abralyt 2000, EasyCap) and conductive electrode cream (Synapse, Med-Tek) were applied to set the electrode impedance to $<5\,\text{k}\Omega$ and to assure long-term stability of the signal with minimal DC potential drifts. The EEG signal was recorded using BrainVision Recorder 1.05 software.

Data processing and analysis

Spreading depolarizations were identified in the subdural recordings by (i) the simultaneous occurrence of a slow potential change in the DC or near-DC frequency range ($<0.05\,\text{Hz}$), and depression of spontaneous activity in the AC frequency range (~0.5–45 Hz) in individual channels; and (ii) the sequential occurrence of slow potential change and depression on adjacent channels, evidencing the propagation of spreading depolarization across the cortex as described previously (Fabricius *et al.*, 2006). Depression durations were scored beginning at the initial decrease in the integral (60 s decay time constant) of the power of the AC-electrocorticography and ending at the start of the recovery phase as described previously (Dreier *et al.*, 2006) (Fig. 1A). The integral of the power is based on a method of computing time integrals with a reset type that resets the integral periodically after a given time, in the manner of decay such as that found in a 'leaky' analogue integrator. This mathematical procedure provides a smoothed curve easing visual assessment of changes in AC-electrocorticography power. The method has become standard to score depression periods of spreading depolarizations (Dreier *et al.*, 2006; Dohmen *et al.*, 2008; Hartings *et al.*, 2011b) and is useful in the screening for ictal epileptic activity (Fabricius *et al.*, 2008; Dreier *et al.*, 2012). In this article, depression periods were scored in each of the five bipolar electrocorticography channels (electrode 1–2, 2–3, 3–4, 4–5 and 5–6) to determine longest and shortest depression period of each spreading depolarization.

Slow potential changes also occurred in some cases when the AC-electrocorticography band was flat, or isoelectric, and therefore lacked depression periods. Baseline isoelectricity may result from non-spreading depression of activity as described previously (Leão, 1947; Dreier, 2011), or from a persistent depression period of a preceding spreading depolarization (Dreier *et al.*, 2006; Fabricius *et al.*, 2006; Dohmen *et al.*, 2008; Oliveira-Ferreira *et al.*, 2010; Hartings *et al.*, 2011b). Slow potential changes occurring in a stereotyped and propagating manner in isoelectric tissue are termed 'silent' spreading depolarizations in the present paper. In a previous paper, the term 'isoelectric' spreading depolarization was used (Hartings *et al.*, 2011b). Both terms refer to the same phenomenon. The term 'isoelectric' spreading depolarization does not imply that the slow potential change remains undetected by the electrocorticography recording.

Apparent propagation velocities were calculated from the DC- or near-DC-electrocorticography signals as the 10-mm separation between subdural electrodes divided by the time interval between the onsets of the slow potential change at adjacent electrodes.

Because of the substantial spatial distance between EEG electrodes at the scalp and subdural electrocorticography electrodes at the brain surface, EEG depressions were accepted as co-registrations in a range of $\pm15\,\text{min}$ around time points of spreading depolarization appearance in the subdural electrodes. Such spatial distance may lead to a substantial time displacement of the co-registration. Although a 30-min window was thus considered for analysis around the electrocorticography starting point of spreading depolarization, the actual delay observed was in general much lower, with a median $<6\,\text{min}$ (see below).

Durations of EEG depressions were quantified in a similar fashion to electrocorticography depressions beginning at the initial decrease in the integral (60 s decay time constant) of the power of the AC-EEG and ending at the start of the recovery phase (Fig. 1A). We arbitrarily considered EEG depressions as significant if a deviation of the integral of power from the baseline (average of 10 min immediately preceding the drop in integral of power) to a level of $\leqslant85\%$ was detected. Amplitudes of the slow potential changes were measured in the DC-EEG from the baseline to the peak negativity (Fig. 1A). Electrocorticography data were analysed by M.W. blinded to the clinical courses and radiological findings. Spreading depolarization time points were then given to C.D. who analysed the EEG data blinded to the durations of the electrocorticography depression periods, the clinical courses and radiological findings. Thereafter, J.D. performed the statistical comparisons between electrocorticography and EEG data provided by M.W. and C.D. blinded to the clinical courses and radiological findings.

Data are given as median (1st, 3rd quartile). Statistical analysis was performed using Mann–Whitney Rank Sum Tests and Spearman's Rank Order Correlations as indicated in the text. $P < 0.05$ was considered statistically significant.

Results

Slow potential changes in scalp and cortical surface recordings of patients with aneurismal subarachnoid haemorrhage

Demographic details are given in Table 1. During 1255.2 h of near-DC/AC-electrocorticography recording time from the cortical
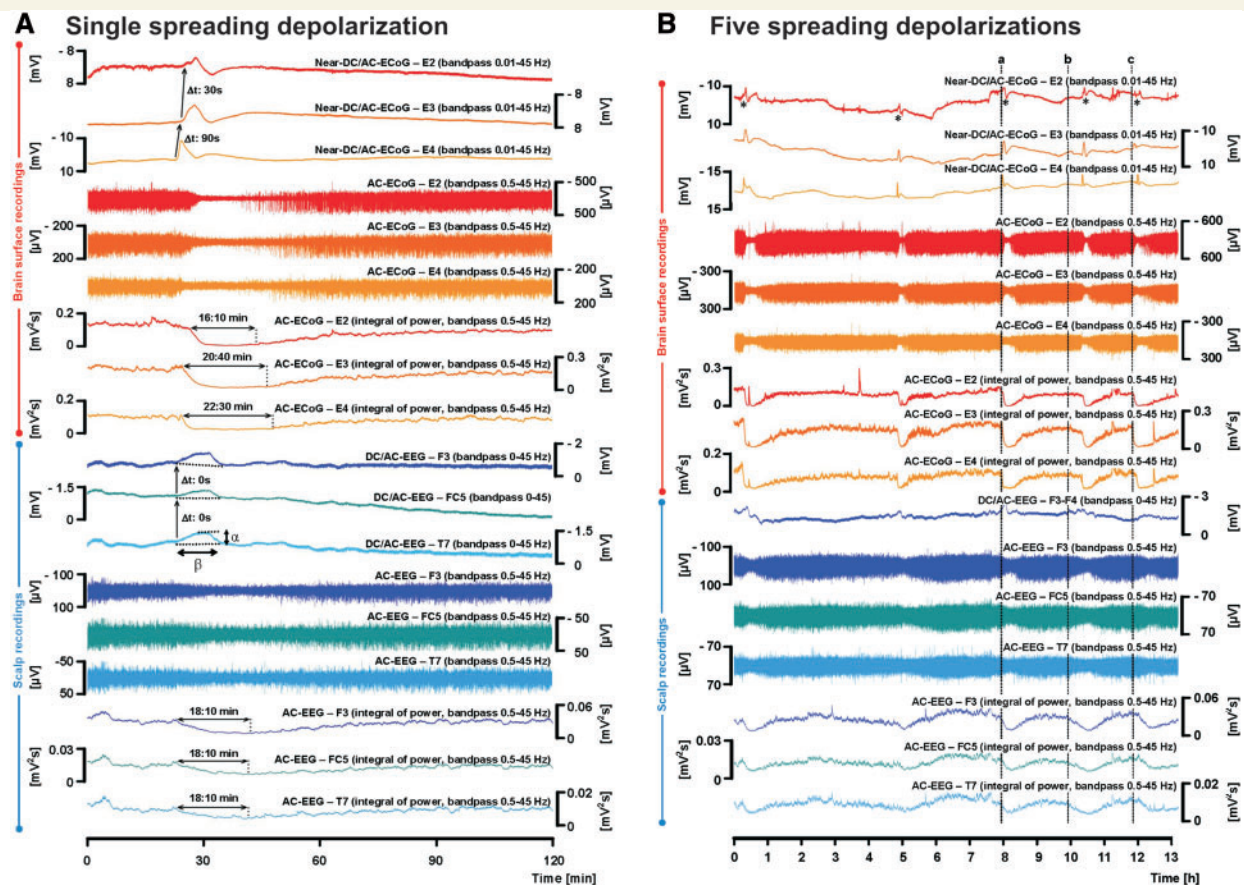
Figure 1 (**A**) Simultaneous recording of spreading depolarization with spreading depression of spontaneous activity in a patient with aneurismal SAH (Case 1 in Table 1) using electrodes at the cortical surface (electrocorticography; ECoG) and scalp EEG. Recordings were performed on Day 5 after aneurismal SAH. Traces 1–9 show the electrocorticography at electrodes E2, E3 and E4 (red, orange and yellow) (subdural electrode strip), whereas traces 10–18 give the EEG at the ipsilateral scalp electrodes F3, FC5 and T7 (dark blue, green and light blue) (international 10–20 electrode system). Traces 1–3 (near-DC/AC-electrocorticography) and traces 10–12 (DC/AC-EEG) display the slow potential change that identifies spreading depolarization. Traces 4–6 (AC-electrocorticography) and 13–15 (AC-EEG) show the associated depression of spontaneous activity in the conventional EEG bandwidth >0.5 Hz. The integral of power of the conventional EEG bandwidth is calculated in traces 7–9 (AC-electrocorticography) and 16–18 (AC-EEG). The figure illustrates how the integral of power is used to score the duration of the depression period from the initial decrease to the start of the recovery phase. Note slow potential change propagation from electrode E4 to E3 to E2 in cortical surface recordings of traces 1–3 (arrows). In contrast, no spread of the slow potential change is identified by visual inspection of the scalp EEG measurements in traces 10–12 (arrows). In similar fashion, propagation of the depression of spontaneous activity is only seen between subdural electrodes (traces 4–9) but not between scalp electrodes (traces 13–18). Moreover, the duration of the depression period is similar at the three scalp electrodes in contrast to subdural recordings where the duration of the depression period differs between electrodes. This inter-regional uniformity of the scalp EEG is due to summation of volume conducted scalp EEG signals from generators widely distributed over the whole hemisphere (see text). Amplitudes of the slow potential changes (α) were measured in the DC-EEG from the baseline (β) to the peak negativity as shown in trace 12. (**B**) Series of five spreading depolarizations (marked by asterisk in trace 1) associated with depression of spontaneous activity recorded by electrocorticography and EEG. The recordings are from the same patient and day as those in (**A**) (delay between **A** and **B**: 8 h and 30 min). Traces 1–9 (red, orange and yellow) show the electrocorticography at subdural electrodes E2, E3 and E4 while traces 10–16 (dark blue, green and light blue) give the EEG at the ipsilateral scalp electrodes. Traces 1–3 (near-DC/AC-electrocorticography) and trace 10 (DC/AC-EEG) display the slow potential changes that identify the spreading depolarizations. Traces 4–6 (AC-electrocorticography) and 11–13 (AC-EEG) show the associated depression of spontaneous activity in the conventional EEG bandwidth >0.5 Hz. The integral of power of the conventional EEG bandwidth is calculated in traces 7–9 (AC-electrocorticography) and 14–16 (AC-EEG). Note the recording time of 13.5 h. Also note that the near-DC-electrocorticography recordings of the slow potential changes at the brain surface indicate that the paths of spreading depolarization in the cortex change from third to fourth to fifth spreading depolarization, so the temporal relationships between electrocorticography and EEG vary between the subsequent spreading depolarizations. Thus, the depression of spontaneous activity of the third spreading depolarization starts almost simultaneously in AC-electrocorticography and AC-EEG (marked by broken line a), whereas the depression of the fourth spreading depolarization starts in the AC-EEG prior to the AC-electrocorticography (marked by broken line b) and the depression of the fifth spreading depolarization starts in the AC-electrocorticography prior to the AC-EEG (marked by broken line c). Similar temporal relationships between cortical surface and scalp also apply to the slow potential changes. Varying paths of spreading depolarizations in the cortex thus translate into slightly varying patterns of slow potential changes and depressions in the scalp DC/AC-EEG.

Table 1 Summary of demographic, treatment and spreading depolarization-related data of the patients

SAH

No.	Age (years), sex	WFNS grade	Fisher grade	Location of aneurysm	Significant proximal vas-spasm	Delayed CT or MRI proven infarct	Intervention	Location of electrode strip	Start of ECoG monitoring (day after insult)	ECoG recording time (h)	No. of SDs in the ECoG	Start of simultaneous ECoG/EEG monitoring (day after insult)	Simultaneous ECoG/EEG recording time (h)	No. of SDs in the simultaneous ECoG/EEG recordings	mRS on Day 14
1	49, F	2	3	PCoA	Yes	Yes	Aneurysm clipping, EVD	Left frontal cortex	0	131.9	38	0	57.5	28	6
2	68, F	4	3	PCoA	Yes	Yes	Aneurysm clipping, EVD	Left frontal cortex	1	258.7	150	1	180.3	53	6
3	48, M	4	3	BA	Yes	No	Aneurysm coiling, EVD	Right frontoparietal cortex	3	269.6	3	3	43.3	3	5
4	63, M	5	3	ACoA	Yes	Yes	Aneurysm coiling and clipping, EVD	Left frontal cortex	0	303.8	16	1	49.8	4	5
5	55, F	4	3	MCA	Yes	Yes	Aneurysm clipping, EVD, hemi-craniectomy	Right frontal cortex	0	291.2	191	1	263.2	187	5

MHS

No.	Age (years), sex	Cause of MHS	Side	territory	Intervention	Location of electrode strip	Start of ECoG monitoring (day after insult)	ECoG recording time (h)	No. of SDs in the ECoG	Start of simultaneous ECoG/EEG monitoring (day after insult)	Simultaneous ECoG/EEG recording time (h)	No. of SDs in the simultaneous ECoG/EEG recordings	mRS on Day 8
6	71, F	ICA occlusion	Left	MCA + ACA	EVD, hemicraniectomy	Left frontal cortex	1	154.6	1	1	65.5	1	5
7	49, F	MCA occlusion	Left	MCA	EVD, hemicraniectomy	Left frontal cortex	1	62.9	24	1	20.1	0	5
8	70, M	MCA occlusion	Right	MCA	EVD, hemicraniectomy	Right frontal cortex	1	176.3	43	1	138.1	18	5
9	54, M	MCA occlusion	Left	MCA	Lysis, EVD, hemicraniectomy	Left frontal cortex	1	81.8	11	1	44.6	1	5

ACA = anterior cerebral artery; ACoA = anterior communicating artery; BA = basilar artery; ECoG = electrocorticography; EVD = extraventricular drainage; F = female; ICA = internal carotid artery; M = male; MCA = middle cerebral artery; mRS = modified Rankin Scale; PCoA = posterior communicating artery; SD = spreading depolarization; WFNS = World Federation of Neurological Surgeons Scale.

Table 2 Comparison between near-DC/AC-electrocorticography and DC/AC-EEG findings in the five patients with aneurismal SAH

Spreading depolarizations in near-DC/AC-ECoG	Proportion of depression periods with a correlate in the AC-EEG	Proportion of SPCs with an SPC correlate in the DC-EEG
Pattern 1: silent spreading depolarizations characterized by persistent depression of spontaneous activity between SPCs ($n = 36$ spreading depolarizations).	Persistent depression of spontaneous activity between SPCs in 2 of 2 patients.	72.2% (SPCs ride on a negative ultraslow potential in 2 of 2 patients).
Pattern 2: spreading depolarizations with depression of spontaneous activity ($n = 239$ spreading depolarizations).	Depression period of spontaneous activity is detected in 46.8% of these spreading depolarizations (34 spreading depolarizations were excluded from this analysis because of an artefact due to the BrainAmp amplifier).	69.9%
	Intervals between successive spreading depolarizations: 44.0 (28.0, 132.0) versus 30.0 (26.5, 51.5) min, detected versus undetected depression periods ($P = 0.001$).	Intervals between successive spreading depolarizations: 33.0 (27.0, 76.5) versus 53.0 (28.0, 130.5) min, detected versus undetected SPCs ($P = 0.009$).

ECoG = electrocorticography; SPC = slow potential change.

surface, the five patients with aneurismal SAH had a total of 398 spreading depolarizations. For further analysis, 694.0 h of simultaneous subdural near-DC/AC-electrocorticography and scalp DC/AC-EEG recording time was available during which 275 of the 398 spreading depolarizations occurred. Slow potential changes of these 275 spreading depolarizations showed a peak-to-peak amplitude of 1.2 (0.8, 1.8) mV (median, quartiles) in the near-DC-electrocorticography recordings; propagation velocity was 5.7 (2.9, 8.2) mm/min assuming an ideal linear spread along the subdural electrode strip. Slow potential change duration was not determined since slow potential change distortion in near-DC recordings precludes assessment of slow potential change duration (Hartings et al., 2009). Subsequently, visual inspection of scalp DC-EEG recordings identified 193 of 275 slow potential changes (70.2%) as illustrated in Figs 1, 2E, 3, 4E and 5. Of note, the intervals between successive spreading depolarizations were significantly shorter, as measured in near-DC-electrocorticography, for spreading depolarizations which displayed the slow potential change in DC-EEG [33.0 (27.0, 76.5) versus 53.0 (28.0, 130.5) min, $P = 0.009$, $n = 273$, Mann–Whitney Rank Sum Test] (Table 2). In other words, DC-EEG was more likely to detect slow potential changes of clustered than isolated spreading depolarizations. Such slow potential changes of clustered, highly frequent spreading depolarizations are shown in Fig. 3B. This observation may indicate that, statistically, larger areas of cortex are simultaneously depolarized in clustered compared with isolated spreading depolarizations.

Median slow potential change amplitude was −272 (−174, −375) µV (range: −65 to −1090 µV) and median slow potential change duration was 5.4 (4.0, 7.1) min in scalp DC-EEG recordings (Fig. 1A). Delay between slow potential change onsets in brain surface near-DC-electrocorticography and scalp DC-EEG recordings was 1.8 (0.8, 3.5) min. Different from subdural electrodes, no spread of the slow potential change was seen between scalp electrodes (Fig. 1A).

The signature of delayed cerebral ischaemia in scalp and cortical surface recordings

Thirty-six of the 275 spreading depolarizations represented silent, clustered spreading depolarizations in the near-DC/AC-electrocorticography recordings. 'Silent' means that spontaneous activity had already ceased before the onset of spreading depolarization. Such silent spreading depolarizations cannot lead to a further clinical deficit in the neurological function represented in the parenchyma undergoing the depolarization as this function was already lost due to the preceding depression of activity (Dreier, 2011; Oliveira-Ferreira et al., 2012). Nevertheless, such silent spreading depolarizations may determine whether this function will be lost permanently as they may damage the neurons irreversibly (Hossmann, 1994; Dreier et al., 2006; Fabricius et al., 2006; Oliveira-Ferreira et al., 2010; Hartings et al., 2011a, b). The persistent depression of spontaneous activity between the silent spreading depolarizations in the AC-electrocorticography was well reflected by the persistent depression of spontaneous activity in the AC-EEG in each case, as illustrated in Fig. 2E. Simultaneously with the silent spreading depolarizations in the near-DC/AC-electrocorticography, DC-EEG displayed slow potential changes riding on a negative ultraslow potential (Fig. 2E).

It has been shown previously that such electrocorticography clusters of recurrent silent spreading depolarizations are associated with delayed ischaemic infarcts after aneurismal SAH (Dreier et al., 2006). Consistently, the cluster of silent spreading depolarizations, demonstrated in Fig. 2E, was associated with a large, new delayed ischaemic infarct in the ipsilateral hemisphere during the recording period, as illustrated in the MRI scans of Fig. 2C1 and C2 compared with those of the preceding MRI shown in Fig. 2B1 and B2. Figure 2A1, A2 and 2D demonstrate localization of subdural recording strip and scalp electrode array, respectively.

In another patient, the subdural DC/AC-electrocorticography was recorded over the infarcting area during infarct evolution in
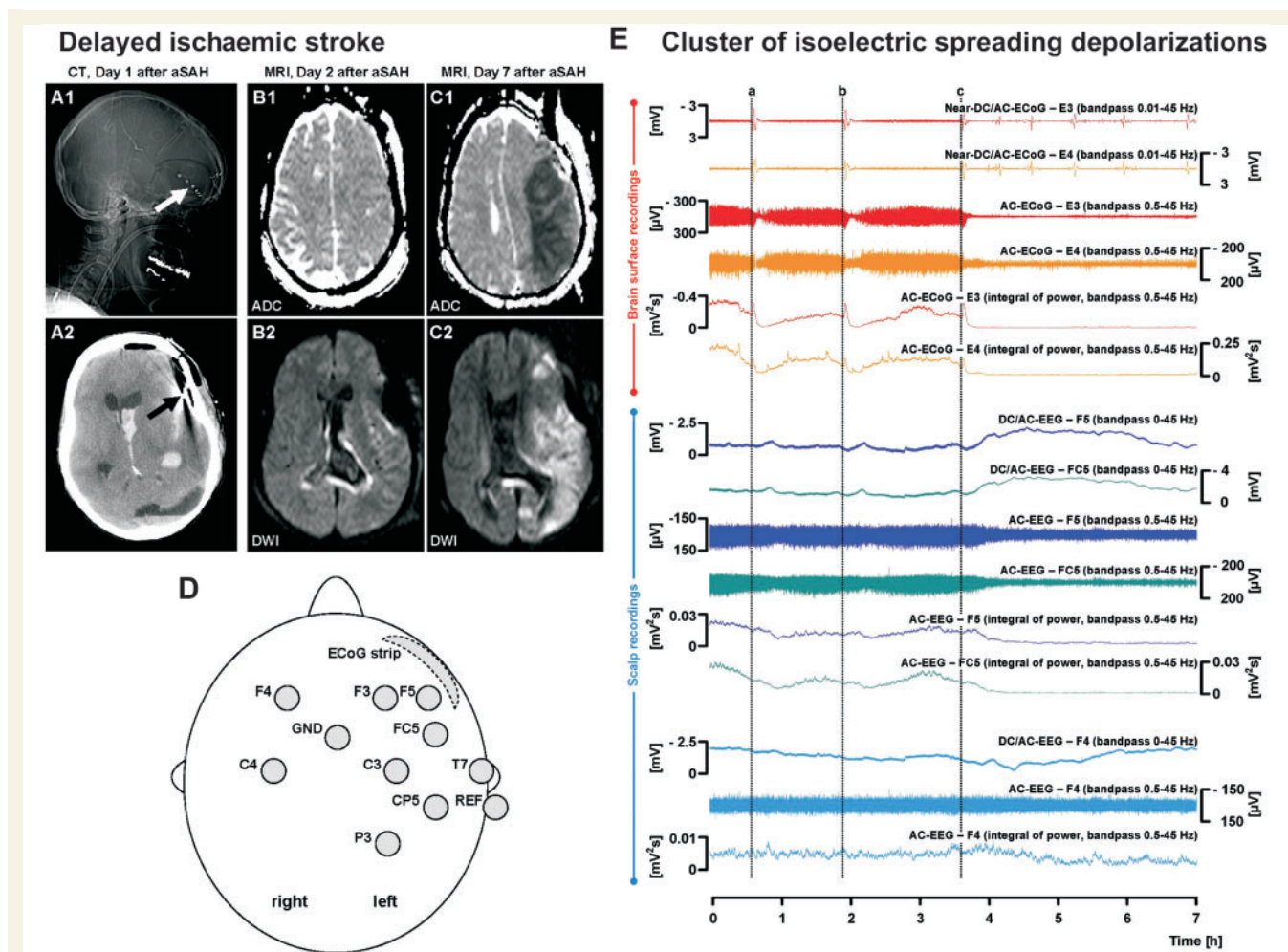
Figure 2 Development of a large delayed ischaemic infarct at the recording area during the monitoring period. (**A1**) The CT scout view shows the orientation of the electrocorticography (EoG) recording strip (marked by white arrow). (**A2**) CT scan showing location of subdural electrode E2 (marked by black arrow). (**B1**) Apparent diffusion coefficient (ADC) map, and (**B2**) diffusion weighted imaging (DWI) of MRI on Day 2 after aneurismal SAH (aSAH). (**C1**) Apparent diffusion coefficient map, and (**C2**) diffusion weighted imaging scan of MRI on Day 7 after aneurismal SAH. Note that the MRI scans of Day 7 display a large new delayed ischaemic infarct in the left middle cerebral artery and posterior cerebral artery territories. (**D**) Scalp electrode array following the 10–20 system, and electrocorticography recording strip. (**E**) Transition from spreading depolarizations associated with depression of spontaneous activity to a cluster of silent spreading depolarizations with persistent depression of activity. The electrocorticography and EEG traces are from the same patient as those in Fig. 1 but only recorded on Day 6 after aneurismal SAH between the two MRIs shown in (**B**) and (**C**). Traces 1–6 give the electrocorticography at electrodes E3 (red) and E4 (orange), and traces 7–12 the EEG at the ipsilateral scalp electrodes F5 (dark blue) and FC5 (green). Traces 13–15 display the EEG at the contralateral scalp electrode F4 (light blue). Traces 1 and 2 (near-DC/AC-electrocorticography) and traces 7 and 8 (DC/AC-EEG) display the slow potential changes that identify the spreading depolarizations. Traces 3 and 4 (AC-electrocorticography) and 9 and 10 (AC-EEG) show the associated depression of spontaneous activity in the conventional EEG bandwidth >0.5 Hz. The integral of power of the conventional EEG bandwidth is calculated in traces 5 and 6 (AC-electrocorticography) and 11 and 12 (AC-EEG). The first two spreading depolarizations during this recording period of 7 h are associated with depression of spontaneous activity followed by recovery (marked by broken lines a and b). The third spreading depolarization (marked by broken line c) initiates the persistent spreading depression of spontaneous activity during which the electrocorticography displays five silent spreading depolarizations ('silent' means that spontaneous activity has already ceased before spreading depolarization onset, see text). Note that the persistent depression of spontaneous AC-electrocorticography activity (traces 3–6) is accompanied by simultaneous depression of spontaneous AC-EEG activity (traces 9–12). Also note the onset of a negative ultraslow potential in scalp electrodes F5 and FC5 (traces 7 and 8) marked by broken line c. In animals, slow potential changes riding on such negative ultraslow potentials are the characteristic signature of neuronal injury (Herreras and Somjen, 1993; Oliveira-Ferreira *et al.*, 2010). Changes at the contralateral electrode F4 are much less pronounced. The mild AC-EEG depression at the contralateral electrode may be caused by the ipsilateral reference at the mastoid. GND = ground; REF = reference.
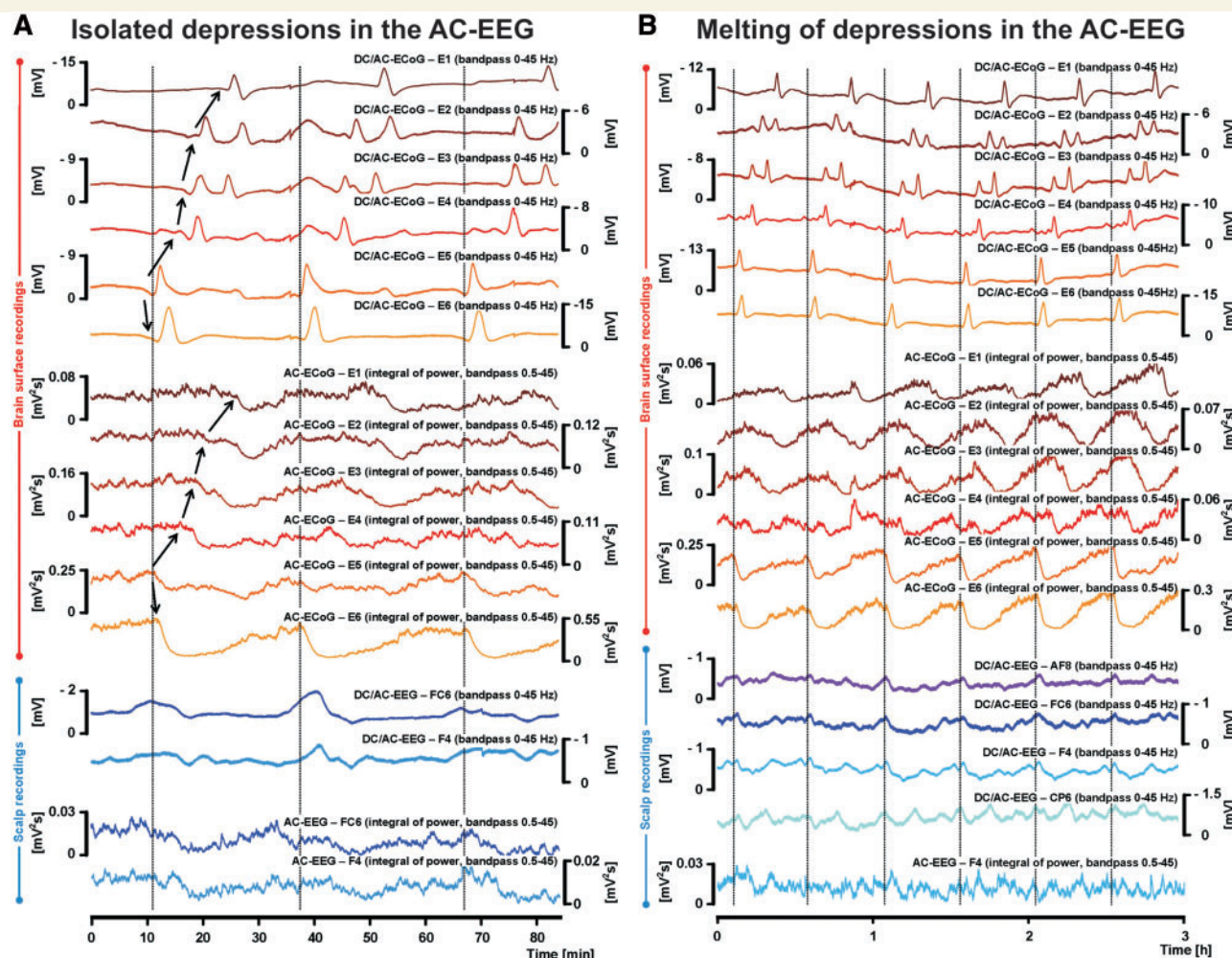
Figure 3 Cluster of repetitive spreading depolarizations in a patient developing a delayed ischaemic infarct remote from the subdural recording strip (Case 5 in Table 1). (**A**) Traces 1–6 display the DC/AC-electrocorticography (ECoG) at subdural electrodes E1–E6. Note the slow potential changes that identify three spreading depolarizations in this episode of 80 min duration (vertical lines). The slow potential changes seem to propagate from electrode E5 to the other electrodes (arrows). Also note large amplitudes of the slow potential changes and slow potential changes with two or even three peaks at electrodes E2 to E5. Such twin peaks could reflect longer depolarizations in deeper layers of the cortex (Herreras and Somjen, 1993). Traces 7–12 give the simultaneous integrals of AC-electrocorticography power demonstrating cycles of spreading depression of spontaneous activity (arrows) followed by recovery. Traces 13 and 14 display the slow potential changes at scalp electrodes FC6 (dark blue) and F4 (light blue) that correspond to the slow potential changes at the cortical surface. The integrals of EEG power in traces 15 (electrode FC6) and 16 (electrode F4) show isolated cycles of depression in spontaneous activity followed by recovery similar to the invasive recordings. In contrast to the subdural recordings, no propagation of slow potential changes and depression periods is observed between scalp electrodes. (**B**) One day later, spreading depolarizations continue to recur at high frequency. Arrangement of traces, electrodes and filter settings is similar to (**A**) but the DC/AC-EEG recordings at scalp electrodes AF8 and CP6 are demonstrated in addition to those at scalp electrodes FC6 and F4 to illustrate variations in scalp slow potential change patterns. Note correspondence between slow potential changes at scalp and cortical surface (vertical lines). It seems that subsequent depression periods are fused at scalp electrode F4 (integral of power in trace 17) although subdural electrodes still show isolated cycles of spreading depression followed by recovery of spontaneous activity at the cortical surface.

addition to the routine near-DC/AC-electrocorticography (Figs 4 and 5). As described previously during infarct evolution in animals (Herreras and Somjen, 1993; Hossmann, 1994; Oliveira-Ferreira et al., 2010), slow potential changes at the cortical surface rode on a characteristic local negative ultraslow injury potential (Fig. 5). DC-EEG recordings simultaneously displayed slow potential

changes riding on a shallow negative ultraslow potential at the scalp. Drop in integral of AC-EEG power, indicating persistent depression of spontaneous activity at the scalp, displayed a similar time course as drop in integral of AC-electrocorticography power, indicating persistent depression of spontaneous activity at the brain surface (Fig. 5).
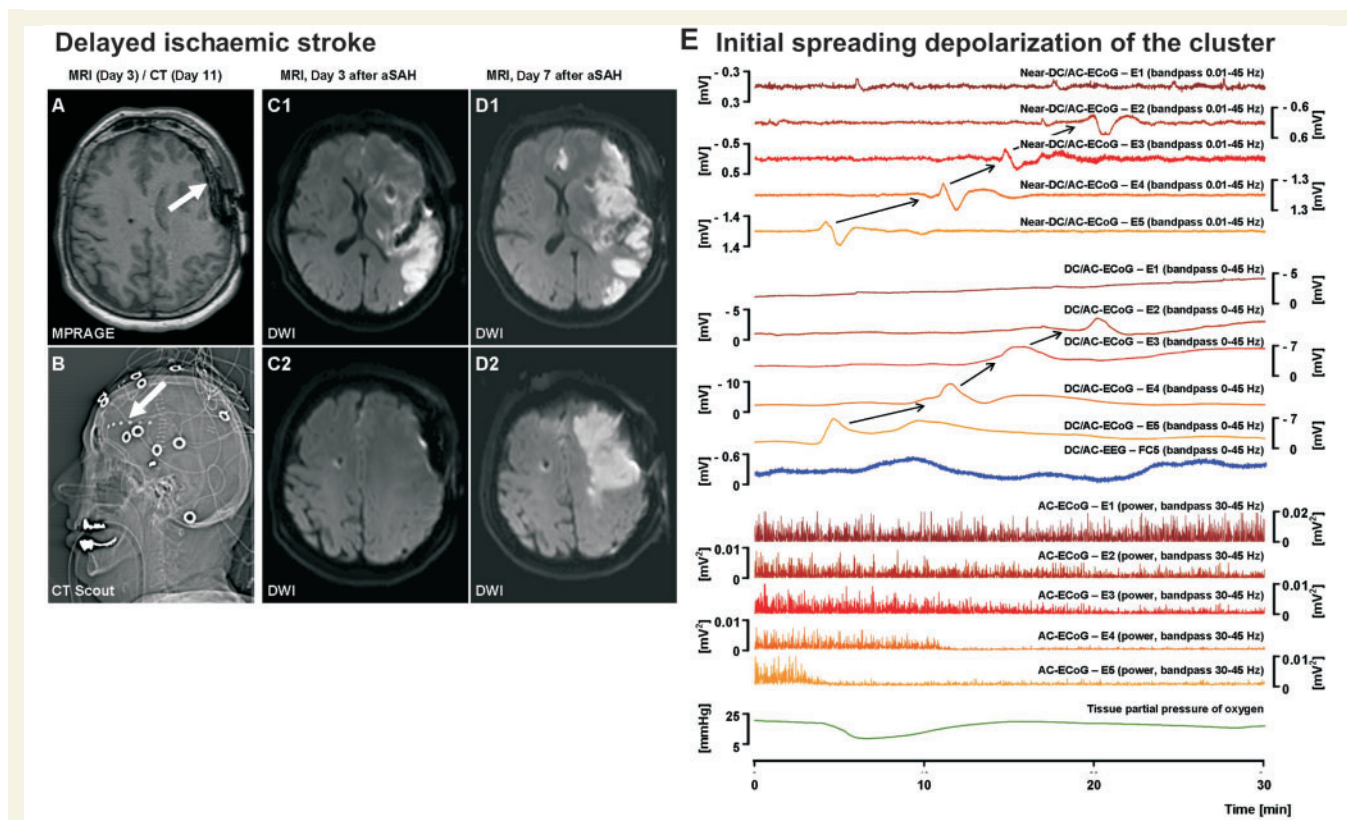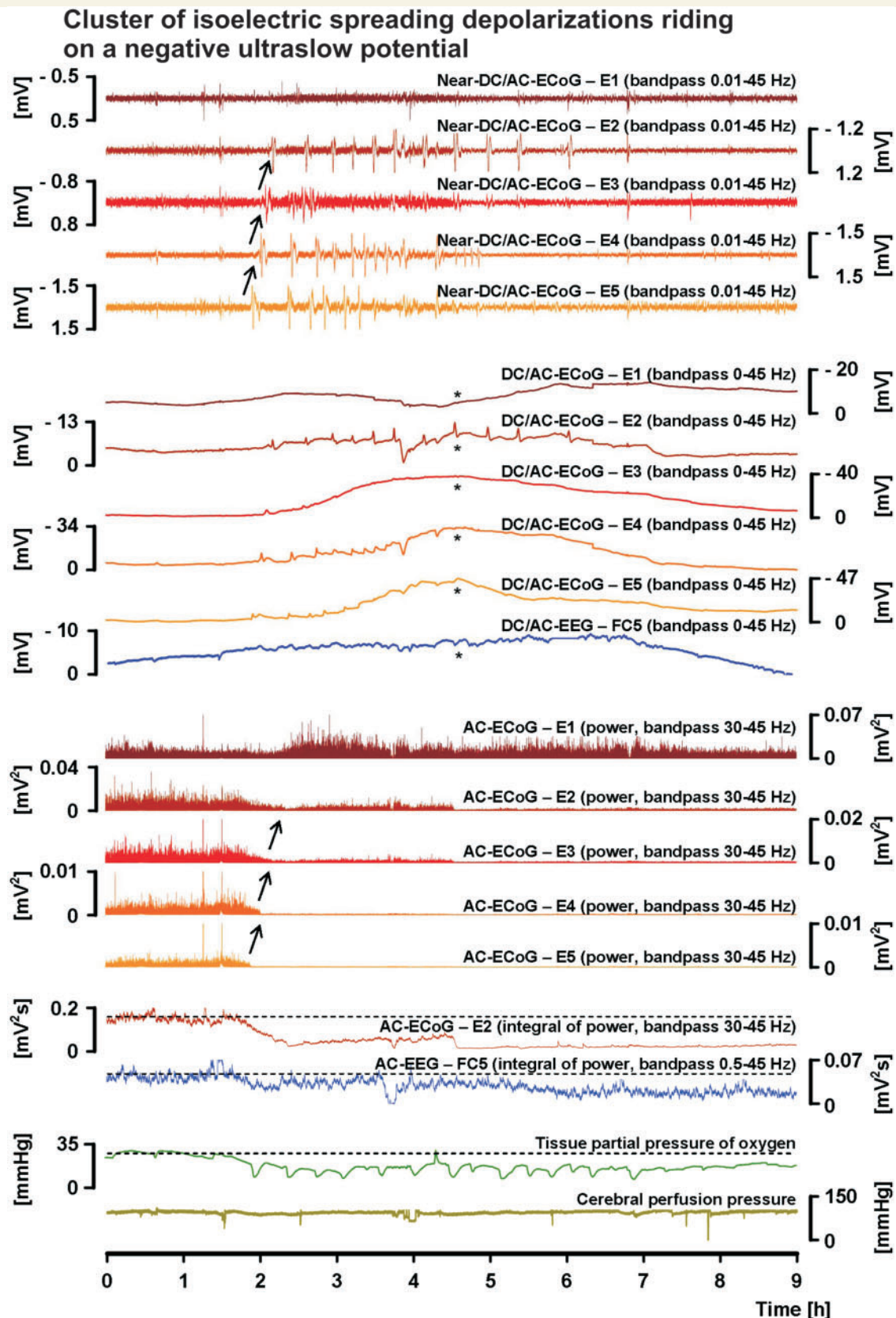
Figure 4 Development of a large delayed ischaemic infarct at the recording area during the monitoring period (Case 2 in Table 1). (**A**) MPRAGE (magnetization prepared rapid gradient echo)-sequence, a T_1-weighted, gradient-echo sequence visualizing the subdural recording strip (marked by white arrow). (**B**) The CT scout view shows the orientation of the electrocorticography (ECoG) recording strip (marked by white arrow). (**C1** and **C2**) Diffusion weighted MRI (DWI) shows an infarct in the posterior territory of the left middle cerebral artery on Day 3. (**D1** and **D2**) On Day 7, a new delayed ischaemic infarct is visualized in the left anterior middle cerebral artery territory including the recording area. Moreover, a small delayed infarct is seen in the left anterior cerebral artery territory. (**E**) The initial spreading depolarization of the cluster is displayed that is completely depicted at lower resolution in Fig. 5. The cluster occurred on Day 4 after aneurismal SAH between the two MRIs of Days 3 and 7. Traces 1–5 show the near-DC/AC-electrocorticography recordings at subdural electrodes E1 to E5 measured by the GT205 amplifier whereas traces 6–10 simultaneously give the DC/AC-electrocorticography recordings measured by the BrainAmp amplifier. Note that the slow potential change is distorted in the near-DC/AC-electrocorticography recordings in traces 2–5, which precludes assessment of its duration (Hartings et al., 2009) in contrast to the slow potential change depicted in the DC/AC-electrocorticography recordings in traces 7–10. The slow potential change propagates from electrode E5 to E2 (arrows). Trace 11 (blue) provides the slow potential change simultaneously measured by electrode FC5 at the scalp. Traces 12–16 depict the spreading depression of spontaneous activity in the power of the AC-electrocorticography at subdural electrodes E1 to E5. The lowest trace (green) displays the tissue partial pressure of oxygen. Abrupt, marked reduction of tissue oxygen accompanies spreading depolarization as measured close to electrode E6 using an intraparenchymal oxygen sensor (Licox, Integra Lifesciences Corporation). This may be the consequence of a combination of reduced blood supply (inverse coupling) with increased oxygen consumption in response to spreading depolarization (Dreier et al., 2009). Note that the spreading depolarization does not propagate to subdural electrode E1. Electrode E1 was positioned on a neighbouring gyrus that was not affected by the new infarct. Because of artefacts in lower frequencies, we chose a bandpass between 30 and 45 Hz to illustrate the depression of spontaneous activity in the subdural recordings.

Isolated depression periods in scalp and cortical surface recordings of patients with aneurismal subarachnoid haemorrhage

In 239 of the 275 spreading depolarizations, near-DC/AC-electrocorticography displayed spreading depression of spontaneous activity. Unfortunately, in 34 of those, depression of spontaneous activity escaped detection in the AC-EEG due to an artefact produced by the automatic drift correction of the BrainAmp amplifier. The automatic drift correction had to be turned on since substantial slow drifts of the EEG signal could occur over the long recording periods. For each of the remaining 205 spreading depolarizations, we identified the AC-electrocorticography channel with the longest and the shortest depression periods using the integral of power of the cortical surface recordings (Fig. 1A). As explained previously, the depression period is an indirect indicator of tissue energy supply since restoration of the spontaneous activity after spreading depolarization is energy dependent (Back et al., 1994; Dreier et al., 2006). In the 205 spreading depolarizations, the median longest depression period

Figure 5 Cluster of silent spreading depolarizations riding on a negative ultraslow potential during development of a new delayed ischaemic infarct (Case 2 in Table 1). Figure 4 depicts the first spreading depolarization of this cluster at high resolution in addition to the neuroimaging findings. Similar to Fig. 4, traces 1–5 show the near-DC/AC-electrocorticography (ECoG) recordings at subdural electrodes E1 to E5 measured by the GT205 amplifier, whereas traces 6–10 simultaneously give the DC/AC-electrocorticography recordings

(continued)

lasted for 12.6 (9.5, 16.8) min and the median shortest depression period for 5.0 (4.1, 8.1) min. During the longest depression period, the integral of power fell to 23.6 (10.5, 35.2)%, and, during the shortest depression period, to 24.2 (14.2, 34.0)%. In only 96 of the 205 spreading depolarizations (46.8%), visual inspection of the AC-EEG identified depression of spontaneous activity. Figure 1 shows examples for such scalp EEG detected depression periods. Proportions of spreading depolarizations with AC-electrocorticography depression that also displayed AC-EEG depression of spontaneous activity ranged from 30% to 75% for the five individual patients with aneurismal SAH. During depression, the integral of scalp AC-EEG power decreased to a median value of 52.5 (36.2, 64.1)% (range: 10.8–82.1%).

Subsequently, we compared the 96 spreading depolarizations where AC-EEG displayed depression periods with the 109 spreading depolarizations where this was not the case. Of note, the interval between successive spreading depolarizations were significantly longer for spreading depolarizations during which AC-EEG displayed a depression period [44.0 (28.0, 132.0) versus 30.0 (26.5, 51.5) min, $P = 0.001$, $n = 205$, Mann–Whitney Rank Sum Test] (Table 2). Moreover, spontaneous activity was depressed to a significantly lower level [integral of AC-electrocorticography power reduction to 15.8 (7.3, 34.7) versus 27.1 (12.5, 35.6)% during the longest depression period, $P = 0.004$, $n = 205$, Mann–Whitney Rank Sum Test]. The EEG reflects a summation of volume-conducted signals from cortical generators widely distributed over the whole hemisphere. These findings thus suggest that highly frequent spreading depolarizations with depression of spontaneous activity in the AC-electrocorticography led to fusion of depression periods in the AC-EEG between subsequent spreading depolarizations. This process is illustrated in Fig. 3A and B.

We then investigated whether the durations of either shortest or longest AC-electrocorticography depression period of spontaneous activity correlated with the duration of the AC-EEG depression period. Both correlated significantly with the AC-EEG depression period ($n = 96$; shortest depression: correlation coefficient: 0.301, $P = 0.003$; longest depression: correlation coefficient: 0.233, $P = 0.023$, Spearman's Rank Order Correlation). We also studied whether the levels to which the spontaneous electrocorticography activity was depressed during the shortest and longest depression periods, respectively, correlated with the level to which the EEG was depressed. Again, significant correlations were found ($n = 96$; shortest depression: correlation coefficient: 0.287, $P = 0.005$; longest depression: correlation coefficient: 0.435, $P < 0.001$, Spearman's Rank Order Correlation). Consistent with the findings for slow potential changes at scalp electrodes, there was no observable spread of AC-EEG depression between scalp electrodes (Fig. 1A). Onset of AC-EEG depression and slow potential change could precede, accompany or succeed onset of AC-electrocorticography depression and slow potential change for different spreading depolarizations of the same patient as shown in Fig. 1B. This change in temporal relationships between brain surface near-DC/AC-electrocorticography and scalp DC/AC-EEG corresponded with different propagation paths in the near-DC/AC-electrocorticography recordings (Fig. 1B). Median delay between AC-electrocorticography and AC-EEG depressions was 5.3 (2.1, 10.1) min.

Patients with malignant hemispheric stroke

Demographic details are given in Table 1. During 475.6 h of near-DC/AC-electrocorticography recording time, the four patients with MHS had a total of 79 spreading depolarizations. For further analysis, 268.2 h of simultaneous near-DC/AC-electrocorticography and DC/AC-EEG recording time was available during which 20 of the 79 spreading depolarizations were observed. The slow potential change of these spreading depolarizations had a peak-to-peak amplitude of 4.5 (3.5, 5.2) mV in the near-DC-electrocorticography and a propagation velocity of 6.1 (3.2, 7.6) mm/min. There was only one silent, clustered spreading depolarization. Hence, 19 spreading depolarizations induced AC-electrocorticography depression of spontaneous activity. Unfortunately, 12 of these were not detected in the scalp DC/AC-EEG due to the artefact produced by the automatic DC drift correction of the BrainAmp amplifier. The remaining seven spreading depolarizations from three of the four patients showed a depression to 50.4 (34.1, 59.1)% similar to the median depression in the patients with aneurismal SAH and lasted for 10.5 (9.4, 22.6) min. The simultaneous shortest and

Figure 5 Continued

measured by the BrainAmp amplifier. The near-DC/AC- and DC/AC-electrocorticography recordings display the slow potential changes that identify the spreading depolarizations. The arrow marks the first spreading depolarization in traces 2–5. The first spreading depolarization causes persistent spreading depression of spontaneous activity, as shown in traces 13–16 (power of subdural electrodes E1–E5, arrows), trace 17 (integral of power at electrode E2) and trace 18 (integral of power at scalp electrode FC5). The multiple subsequent spreading depolarizations in traces 2–5 and 7–10, respectively, occur during this persistent depression of spontaneous activity (silent spreading depolarizations). Note in the DC/AC-electrocorticography recordings of traces 7–10 that the recurrent slow potential changes ride on a negative ultraslow potential (marked by the stars). This negative ultraslow injury potential is largest at electrodes E3 to E5 and seems reflected in a shallow negative ultraslow potential at scalp electrode FC5 (trace 11). Note that no slow potential changes/spreading depolarizations occur at subdural electrode E1 which was located on another gyrus spared from infarction. Interestingly, an ultraslow positive potential (current source) is seen at subdural electrode E1 (star at trace 6) in contrast to the ultraslow negative potential (current sink) at the other electrodes, remarkably similar to findings in and around infarcts in animals (Oliveira-Ferreira *et al.*, 2010). Trace 19 shows a persistent decrease of tissue partial pressure of oxygen as measured with an oxygen sensor. Note recurrent decreases of tissue oxygen in response to the spreading depolarizations. Cerebral perfusion pressure is constant during the event. Because of artefacts in lower frequencies, we chose a bandpass between 30 and 45 Hz to illustrate the depression of spontaneous activity in the subdural recordings.

longest AC-electrocorticography depression periods lasted for 7.7 (6.4, 15.3) and 2.8 (2.3, 3.7) min, respectively; the integral of AC-electrocorticography power was depressed to 24.7 (17.9, 34.4) and 29.7 (21.4, 49.5)%, respectively. In the scalp DC-EEG recordings, median slow potential change amplitude was −305 (range: −107, −517) μV and median slow potential change duration was 7.4 (4.4, 8.3) min. Correlations between electrocorticography and EEG parameters were not analysed because of insufficient statistical power.

Discussion

It has been known for a long time that cerebral ischaemia is associated with marked changes in the human scalp AC-EEG, such as polymorphic delta activity, focal attenuation as well as loss of fast activity and sleep spindles (Cohn *et al.*, 1948). These changes in spontaneous activity were first used diagnostically for the intraoperative monitoring during carotid endarterectomy (Sharbrough *et al.*, 1973). Later, a number of approaches using scalp AC-EEG have been developed to detect the advent of delayed cerebral ischaemia in patients with aneurismal SAH (Labar *et al.*, 1991; Rivierez *et al.*, 1991; Vespa *et al.*, 1997; Claassen *et al.*, 2004, 2005). Clinical application of quantitative AC-EEG technology was tested in different studies and was found clinically useful in the screening for delayed cerebral ischaemia. The best quantitative AC-EEG parameter for this purpose has remained controversial. Ratios of fast over slow activity and trend analysis of total power, in similar fashion to our study, are among the favoured approaches (Labar *et al.*, 1991; Vespa *et al.*, 1997; Claassen *et al.*, 2005). It is seen as a strength of scalp AC-EEG that the changes associated with delayed cerebral ischaemia are widespread so these changes provide a summary measure for very different locations of the cerebrum that can be affected by delayed cerebral ischaemia. Use at the bedside is another obvious advantage of scalp EEG. On the other hand, there are some disadvantages: artefacts by scalp electrodes may confound neuroimaging studies (Claassen *et al.*, 2005). Moreover, interpretation of quantitative AC-EEG parameters has been only recommended with caution (Claassen *et al.*, 2005). They should not be interpreted in isolation but in combination with the underlying raw AC-EEG by a person trained in this analysis since a multitude of artefacts as well as extracranial factors such as scalp swelling may confound them. Furthermore, labour intensity is a problem for continuous EEG recording, an EEG technician being constantly needed to ensure high-quality measurements and neurophysiologists having to evaluate enormous amounts of EEG data.

Classic diagnostic tools for detection of ischaemic lesion progression

Quantitative AC-EEG in isolation is not sufficient for the diagnosis of delayed cerebral ischaemia but it serves as a screening tool (Claassen *et al.*, 2005). This implies that additional studies are currently needed to confirm the diagnosis of delayed cerebral ischaemia such as clinical examination, digital subtraction angiography, transcranial Doppler sonography or neuroimaging (Claassen

et al., 2005). Unfortunately, these confirmatory studies have limitations as well: clinical examinations are of limited value in stuporous or comatose patients with aneurismal SAH. Digital subtraction angiography remains the gold standard for the diagnosis of proximal vasospasm but is not without risk for the patient, and the value of digital subtraction angiography for the diagnosis of delayed cerebral ischaemia has been increasingly questioned in recent years (Vergouwen *et al.*, 2010). First, there is now clear evidence from autopsy and neuroimaging studies that delayed cerebral ischaemia can occur without angiographic vasospasm (Neil-Dwyer *et al.*, 1994; Dreier *et al.*, 2002; Weidauer *et al.*, 2008; Woitzik *et al.*, 2011). Moreover, in contrast to a significant association between unfavourable clinical outcome and delayed cerebral ischaemia (Vergouwen *et al.*, 2011), in a recent meta-analysis, no association was found between unfavourable outcome and proximal vasospasm (Etminan *et al.*, 2011). The validity of transcranial Doppler-sonography for the diagnosis of delayed cerebral ischaemia is even more restricted since, as a surrogate method for digital subtraction angiography, significant correspondence between digital subtraction angiography and transcranial Doppler-sonography was only found for relatively small ranges of transcranial Doppler-sonography mean velocities in the middle cerebral arteries <120 and >200 cm/s (Vora *et al.*, 1999). CT is a reliable tool to detect delayed territorial infarcts but lesion maturation takes several hours before CT reliably identifies an infarct and so diagnosis is delayed. Moreover, cortical infarcts are the predominant pathomorphological correlate of delayed cerebral ischaemia in autopsy studies but frequently escape detection by CT (Neil-Dwyer *et al.*, 1994; Dreier *et al.*, 2002; Weidauer *et al.*, 2008). Furthermore, with few exceptions, CT is not a bedside tool but requires transport to the scanner tying up human resources from the hospital. MRI is the gold standard for the detection of delayed ischaemic stroke since its sensitivity for small lesions is significantly higher compared with that of CT (Shimoda *et al.*, 2001; Dreier *et al.*, 2002; Vergouwen *et al.*, 2010). Nevertheless, mild ischaemia without structural injury still escapes detection by MRI. Moreover, MRI shares with CT the transport problem of the patient between scanner and intensive care unit. Other methods of complementary value are imaging techniques assessing cerebral blood flow but their practical use is limited by the enormous temporal dynamics of cerebral blood flow that can drop to ischaemic levels within seconds and change to hyperperfusion within minutes up to hours thereafter as observed in a characteristic fashion using continuous subdural laser-Doppler flowmetry in patients with aneurismal SAH (Dreier *et al.*, 2009). This behaviour corresponds well with the marked diversity of cerebral blood flow patterns in imaging studies of delayed cerebral ischaemia (Minhas *et al.*, 2003).

Continuous quantitative neuromonitoring at the bedside for detection of ischaemic lesion progression

The combination of the invasive neuromonitoring tools such as subdural DC/AC-electrocorticography, tissue partial pressure of

oxygen measurements, laser-Doppler flowmetry of regional cerebral blood flow and slow and rapid sampling microdialysis now allow for the assessment of early and delayed pathophysiology in patients with aneurismal SAH and MHS in practically the same detail as in animal studies (Dreier *et al.*, 2009; Bosche *et al.*, 2010; Feuerstein *et al.*, 2010; Oliveira-Ferreira *et al.*, 2010). Cut-off values for these methods are currently being developed to guide treatment allocation in patients at risk for delayed cerebral ischaemia (Dreier, 2011). However, such cut-off values could be limited by the more or less restricted sample volumes of the invasive tools. The largest sample volume may be that of DC/AC-electrocorticography since spreading depolarizations invade the tissue surrounding the ischaemic zone and high recurrence rates may indicate even remotely developing ischaemic lesions (Dreier *et al.*, 2006; Dohmen *et al.*, 2008; Oliveira-Ferreira *et al.*, 2010). However, even if reliable cut-off values are calculated for early diagnosis of delayed cerebral ischaemia after aneurismal SAH or ischaemic lesion progression after MHS, a slight risk for local infection or haemorrhage will remain with invasive probes (Espinosa *et al.*, 1994; Lee *et al.*, 2000). This obstacle will limit probe implantation to patients requiring neurosurgical interventions. Therefore, the ultimate goal for diagnostic development is powerful, non-invasive recording technology for use at the bedside that has been validated by comparison with invasive technology. The present study represents a significant first step in this process as our invasive near-DC- and DC/AC-electrocorticography recordings have demonstrated the pathophysiological basis of the previously described scalp AC-EEG changes in the course of aneurismal SAH and ischaemic stroke. Our findings thus indicate that the characteristic loss of AC-EEG power associated with ischaemia (Labar *et al.*, 1991; Vespa *et al.*, 1997; Claassen *et al.*, 2004) is caused by clusters of spreading depolarizations associated with the depression of spontaneous activity.

In addition, we have identified two other, promising signals while recording the DC-component of the scalp EEG. It was not believed possible to record the slow potential change, the extracellular index of spreading depolarization, in scalp DC-EEG recordings as the potent capacitive resistance of dura and skull would filter the slow voltage variation (Dreier, 2011). In the present study, the large majority of slow potential changes in the near-DC- and DC-electrocorticography were nevertheless accompanied by slow potential changes in the scalp DC-EEG. Furthermore, in two cases, scalp DC-EEG recordings were performed during early infarct evolution, and scalp electrodes were placed over the infarcting area. In these cases, scalp DC-EEG recorded a negative ultraslow potential, the classical extracellular index of neuronal injury in animal experiments (Herreras and Somjen, 1993; Lehmenkühler *et al.*, 1999; Oliveira-Ferreira *et al.*, 2010). In one of these cases, DC-electrocorticography was recorded at the cortical surface in addition to the routine near-DC-electrocorticography while the subdural electrode strip overlaid the region of the developing infarct. This allowed us to measure in parallel the negative ultraslow injury potential at both scalp and cortical surface. Similar to recordings in rats, the DC-shifts of spreading depolarizations did not reverse between cortical surface and scalp as these potentials are generated in the parenchyma rather than at the interface between blood and

brain (Lehmenkühler *et al.*, 1991). However, future experimental studies should address whether and how the signals are influenced by the fresh craniotomy in the patients with aneurismal SAH or by the decompressive hemicraniectomy in the patients with MHS.

Visual inspection did not detect a spread of slow potential change or AC-EEG depression of spontaneous activity between different scalp electrodes. This is likely explained by the fact that the scalp EEG is influenced by volume conduction from many superposed sources of the whole hemisphere. For spread reconstruction, disentanglement of cortical generators from scalp DC/AC-EEG recordings would require more complex mathematical procedures such as virtual source montage or principal components analysis (Miller *et al.*, 2007).

Conclusion and future goals

The strength of the combined bedside recording of the scalp ultraslow potential, slow potential change and depression of spontaneous activity is that they potentially allow for the instantaneous, on-line detection of ischaemic injury onset and progression in a large patient population. This would allow for targeted treatment to begin earlier than with diagnosis based on any imaging modality, which requires time for lesion maturation as well as time for patient transport to the scanner. To come to this point, the electrophysiological techniques require further development and careful analysis of limitations. In principle, the negative ultraslow potential of ischaemic injury is the largest electrophysiological signal at the human brain surface (Leão, 1947; Oliveira-Ferreira *et al.*, 2010), even larger than the slow potential change, which can reach up to 25 mV (Dreier *et al.*, 2009; Oliveira-Ferreira *et al.*, 2010) and much larger than the DC potential associated with epileptic seizure activity, which may reach up to 2 mV at the cortical surface (Dreier *et al.*, 2012) and 30–150 μV at the scalp (Miller *et al.*, 2007). However, the DC potential can be confounded by many different generators including the eyeball, tongue, blood–brain barrier and the skin (Miller *et al.*, 2007). The galvanic skin response at the scalp can be avoided by slight puncturing of the skin epithelia during electrode application. Other causes of DC potentials such as movements, jugular vein compression or chemical factors like changes in carbon dioxide tension, are more difficult to control, and not all DC potential generators may be known.

Further development of the techniques applied here could make it possible in the future to alert the neurointensive care specialist of the onset or progression of neuronal injury in a fashion similar to the use of continuous electrocardiography to detect cardiac arrhythmia. A number of research goals have to be achieved for this purpose. Thus, it is necessary to calculate sensitivity and specificity for a cut-off value of duration in spreading depolarization-induced depression of spontaneous activity measured by electrocorticography or EEG that indicates delayed ischaemic stroke after aneurismal SAH or ischaemic lesion progression in MHS, lesion progression being assessed using serial MRI (Dreier, 2011). For this analysis, recording of slow potential changes would serve the differentiation between spreading depolarization-induced depression and other types of depression

such as depression of spontaneous activity by sedatives for example. Moreover, a better understanding of the complex generators and artefacts underlying DC potential changes at the brain surface and scalp is needed to make full use of slow potential change and ultraslow potential. Polymer researchers should develop electrodes with similar low-frequency recording properties as Ag/AgCl electrodes but without their toxicity to replace the platinum electrodes for the subdural recordings. Platinum has much better low-frequency recording properties than stainless steel but is polarizable and thus inferior to Ag/AgCl (Tallgren et al., 2005). Mathematical tools such as virtual source montage and principal components analysis should be applied to identify the characteristic propagation of spreading depolarizations using scalp DC/AC-EEG in analogous fashion to the source localization of epileptic seizure activity (Miller et al., 2007). The spread would add another criterion to distinguish spreading depolarizations from other bioelectrical phenomena and artefacts. Electrocorticography recordings at the brain surface could be used to validate such mathematical tools since they identify with high accuracy sources of slow potential changes. Additional criteria for the differential diagnosis of electrophysiological signals could be derived from non-invasive surrogate measures of cerebral blood flow such as near-infrared or diffuse correlation spectroscopy that can be applied at the bedside (Obrig and Villringer, 2003; Durduran et al., 2010). All this should be flanked by the development of software packages and hardware for automated, on-line analysis at the bedside on the intensive care unit. We believe that the findings of the present study are promising for this development since, in all patients, even visual inspection of the raw time-compressed DC/AC-EEG data was sufficient to identify clear reflections of the spreading depolarizations at the scalp.

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References

Back T, Kohno K, Hossmann KA. Cortical negative DC deflections following middle cerebral artery occlusion and KCl-induced spreading depression: effect on blood flow, tissue oxygenation, and electroencephalogram. J Cereb Blood Flow Metab 1994; 14: 12–9.

Bosche B, Graf R, Ernestus RI, Dohmen C, Reithmeier T, Brinker G, et al. Recurrent spreading depolarizations after SAH decrease oxygen availability in human cerebral cortex. Ann Neurol 2010; 67: 607–17.

Canals S, Makarova I, Lopez-Aguado L, Largo C, Ibarz JM, Herreras O. Longitudinal depolarization gradients along the somatodendritic axis of CA1 pyramidal cells: a novel feature of spreading depression. J Neurophysiol 2005; 94: 943–51.

Claassen J, Hirsch LJ, Kreiter KT, Du EY, Connolly ES, Emerson RG, et al. Quantitative continuous EEG for detecting delayed cerebral ischemia in patients with poor-grade subarachnoid hemorrhage. Clin Neurophysiol 2004; 115: 2699–710.

Claassen J, Mayer SA, Hirsch LJ. Continuous EEG monitoring in patients with subarachnoid hemorrhage. J Clin Neurophysiol 2005; 22: 92–8.

Cohn R, Raines GN, Mulder DW, Neumann MA. Cerebral vascular lesions; electroencephalographic and neuropathologic correlations. Arch Neurol Psychiatry 1948; 60: 165–81.

Dohmen C, Sakowitz OW, Fabricius M, Bosche B, Reithmeier T, Ernestus RI, et al. Spreading depolarizations occur in human ischemic stroke with high incidence. Ann Neurol 2008; 63: 720–8.

Donnan GA. The 2007 Feinberg lecture: a new road map for neuroprotection. Stroke 2008; 39: 242.

Dreier JP. The role of spreading depression, spreading depolarization and spreading ischemia in neurological disease. Nat Med 2011; 17: 439–47.

Dreier JP, Major S, Manning A, Woitzik J, Drenckhahn C, Steinbrink J, et al. Cortical spreading ischaemia is a novel process involved in ischaemic damage in patients with aneurysmal subarachnoid haemorrhage. Brain 2009; 132: 1866–81.

Dreier JP, Major S, Pannek HW, Woitzik J, Scheel M, Wiesenthal D, et al. Spreading convulsions, spreading depolarization, and epileptogenesis in human cerebral cortex. Brain 2012; 135: 259–76.

Dreier JP, Sakowitz OW, Harder A, Zimmer C, Dirnagl U, Valdueza JM, et al. Focal laminar cortical MR signal abnormalities after subarachnoid hemorrhage. Ann Neurol 2002; 52: 825–9.

Dreier JP, Woitzik J, Fabricius M, Bhatia R, Major S, Drenckhahn C, et al. Delayed ischaemic neurological deficits after subarachnoid haemorrhage are associated with clusters of spreading depolarizations. Brain 2006; 129: 3224–37.

Durduran T, Choe R, Baker WB, Yodh AG. Diffuse optics for tissue monitoring and tomography. Rep Progr Phys 2010; 73: 076701.

Espinosa J, Olivier A, Andermann F, Quesney F, Dubeau F, Savard G. Morbidity of chronic recording with intracranial depth electrodes in 170 patients. Stereotact Funct Neurosurg 1994; 63: 63–5.

Etminan N, Vergouwen MD, Ilodigwe D, Macdonald RL. Effect of pharmaceutical treatment on vasospasm, delayed cerebral ischemia, and clinical outcome in patients with aneurysmal subarachnoid hemorrhage: a systematic review and meta-analysis. J Cereb Blood Flow Metab 2011; 31: 1443–51.

Fabricius M, Fuhr S, Bhatia R, Boutelle M, Hashemi P, Strong AJ, et al. Cortical spreading depression and peri-infarct depolarization in acutely injured human cerebral cortex. Brain 2006; 129: 778–90.

Fabricius M, Fuhr S, Willumsen L, Dreier JP, Bhatia R, Boutelle MG, et al. Association of seizures with cortical spreading depression and peri-infarct depolarisations in the acutely injured human brain. Clin Neurophysiol 2008; 119: 1973–84.

Feuerstein D, Manning A, Hashemi P, Bhatia R, Fabricius M, Tolias C, et al. Dynamic metabolic response to multiple spreading depolarizations in patients with acute brain injury: an online microdialysis study. J Cereb Blood Flow Metab 2010; 30: 1343–55.

Hartings JA, Bullock MR, Okonkwo DO, Murray LS, Murray GD, Fabricius M, et al. Spreading depolarisations and outcome after traumatic brain injury: a prospective observational study. Lancet Neurol 2011a; 10: 1058–64.

Hartings JA, Strong AJ, Fabricius M, Manning A, Bhatia R, Dreier JP, et al. Spreading depolarizations and late secondary insults after traumatic brain injury. J Neurotrauma 2009; 26: 1857–66.

Hartings JA, Watanabe T, Dreier JP, Major S, Vendelbo L, Fabricius M. Recovery of slow potentials in AC-coupled electrocorticography: application to spreading depolarizations in rat and human cerebral cortex. J Neurophysiol 2009; 102: 2563–75.

Hartings JA, Watanabe T, Bullock MR, Okonkwo DO, Fabricius M, Woitzik J, et al. Spreading depolarizations have prolonged direct current shifts and are associated with poor outcome in brain trauma. Brain 2011b; 134: 1529–40.

Herreras O, Somjen GG. Analysis of potential shifts associated with recurrent spreading depression and prolonged unstable spreading depression induced by microdialysis of elevated K+ in hippocampus of anesthetized rats. Brain Res 1993; 610: 283–94.

Hossmann KA. Viability thresholds and the penumbra of focal ischemia. Ann Neurol 1994; 36: 557–65.

Juttler E, Bosel J, Amiri H, Schiller P, Limprecht R, Hacke W, et al. DESTINY II: DEcompressive Surgery for the Treatment of malignant INfarction of the middle cerebral arterY II. Int J Stroke 2011; 6: 79–86.

Kistler JP, Crowell RM, Davis KR, Heros R, Ojemann RG, Zervas T, et al. The relation of cerebral vasospasm to the extent and location of subarachnoid blood visualized by CT scan: a prospective study. Neurology 1983; 33: 424–36.

Klatzo I. Pathophysiological aspects of brain edema. Acta Neuropathol 1987; 72: 236–9.

Klem GH, Luders HO, Jasper HH, Elger C. The ten-twenty electrode system of the International Federation. The International Federation of Clinical Neurophysiology. Electroencephalogr Clin Neurophysiol Suppl 1999; 52: 3–6.

Labar DR, Fisch BJ, Pedley TA, Fink ME, Solomon RA. Quantitative EEG monitoring for patients with subarachnoid hemorrhage. Electroencephalogr Clin Neurophysiol 1991; 78: 325–32.

LaManna JC, Rosenthal M. Effect of ouabain and phenobarbital on oxidative metabolic activity associated with spreading cortical depression in cats. Brain Res 1975; 88: 145–9.

Leão AAP. Further observations on the spreading depression of activity in the cerebral cortex. J Neurophysiol 1947; 10: 409–14.

Lee WS, Lee JK, Lee SA, Kang JK, Ko TS. Complications and results of subdural grid electrode implantation in epilepsy surgery. Surg Neurol 2000; 54: 346–51.

Lehmenkühler A, Richter F, Poppelmann T. Hypoxia- and hypercapnia-induced DC potential shifts in rat at the scalp and the skull are opposite in polarity to those at the cerebral cortex. Neurosci Lett 1999; 270: 67–70.

Lehmenkühler A, Richter F, Scheller D, Speckmann EJ. Non-invasive DC recordings from the skull and the skin during cortical spreading depression: a model of detection of migraine. In: Olesen J, editor. Frontiers in headache research Migraine and other headaches: The vascular mechanisms. New York: Raven Press; 1991. p. 167–70.

Lo EH. A new penumbra: transitioning from injury into repair after stroke. Nat Med 2008; 14: 497–500.

Miller JW, Kim W, Holmes MD, Vanhatalo S. Ictal localization by source analysis of infraslow activity in DC-coupled scalp EEG recordings. Neuroimage 2007; 35: 583–97.

Minhas PS, Menon DK, Smielewski P, Czosnyka M, Kirkpatrick PJ, Clark JC, et al. Positron emission tomographic cerebral perfusion disturbances and transcranial Doppler findings among patients with neurological deterioration after subarachnoid hemorrhage. Neurosurgery 2003; 52: 1017–22 discussion 22–4.

Nakamura H, Strong AJ, Dohmen C, Sakowitz OW, Vollmar S, Sue M, et al. Spreading depolarizations cycle around and enlarge focal ischaemic brain lesions. Brain 2010; 133: 1994–2006.

Neil-Dwyer G, Lang DA, Doshi B, Gerber CJ, Smith PW. Delayed cerebral ischaemia: the pathological substrate. Acta Neurochir (Wien) 1994; 131: 137–45.

Obrig H, Villringer A. Beyond the visible–imaging the human brain with light. J Cereb Blood Flow Metab 2003; 23: 1–18.

Oliveira-Ferreira AI, Milakara D, Alam M, Jorks D, Major S, Hartings JA, et al. Experimental and preliminary clinical evidence of an ischemic zone with prolonged negative DC shifts surrounded by a normally perfused tissue belt with persistent electrocorticographic depression. J Cereb Blood Flow Metab 2010; 30: 1504–19.

Oliveira-Ferreira AI, Winkler MKL, Reiffurth C, Milakara D, Woitzik J, Dreier JP. Spreading depolarization, a pathophysiological mechanism of stroke and migraine aura. Future Neurol 2012; 7: 45–64.

Risher WC, Ard D, Yuan J, Kirov SA. Recurrent spontaneous spreading depolarizations facilitate acute dendritic injury in the ischemic penumbra. J Neurosci 2010; 30: 9859–68.

Rivierez M, Landau-Ferey J, Grob R, Grosskopf D, Philippon J. Value of electroencephalogram in prediction and diagnosis of vasospasm after intracranial aneurysm rupture. Acta Neurochir (Wien) 1991; 110: 17–23.

Sharbrough FW, Messick JM Jr, Sundt TM Jr. Correlation of continuous electroencephalograms with cerebral blood flow measurements during carotid endarterectomy. Stroke 1973; 4: 674–83.

Shimoda M, Takeuchi M, Tominaga J, Oda S, Kumasaka A, Tsugane R. Asymptomatic versus symptomatic infarcts from vasospasm in patients with subarachnoid hemorrhage: serial magnetic resonance imaging. Neurosurgery 2001; 49: 1341–8 discussion 8–50.

Somjen GG. Mechanisms of spreading depression and hypoxic spreading depression-like depolarization. Physiol Rev 2001; 81: 1065–96.

Strong AJ, Fabricius M, Boutelle MG, Hibbins SJ, Hopwood SE, Jones R, et al. Spreading and synchronous depressions of cortical activity in acutely injured human brain. Stroke 2002; 33: 2738–43.

Takano T, Tian GF, Peng W, Lou N, Lovatt D, Hansen AJ, et al. Cortical spreading depression causes and coincides with tissue hypoxia. Nat Neurosci 2007; 10: 754–62.

Tallgren P, Vanhatalo S, Kaila K, Voipio J. Evaluation of commercially available electrodes and gels for recording of slow EEG potentials. Clin Neurophysiol 2005; 116: 799–806.

van Gijn J, Rinkel GJ. Subarachnoid haemorrhage: diagnosis, causes and management. Brain 2001; 124: 249–78.

Vergouwen MD, Etminan N, Ilodigwe D, Macdonald RL. Lower incidence of cerebral infarction correlates with improved functional outcome after aneurysmal subarachnoid hemorrhage. J Cereb Blood Flow Metab 2011; 31: 1545–53.

Vergouwen MD, Vermeulen M, van Gijn J, Rinkel GJ, Wijdicks EF, Muizelaar JP, et al. Definition of delayed cerebral ischemia after aneurysmal subarachnoid hemorrhage as an outcome event in clinical trials and observational studies: proposal of a multidisciplinary research group. Stroke 2010; 41: 2391–5.

Vespa PM, Nuwer MR, Juhasz C, Alexander M, Nenov V, Martin N, et al. Early detection of vasospasm after acute subarachnoid hemorrhage using continuous EEG ICU monitoring. Electroencephalogr Clin Neurophysiol 1997; 103: 607–15.

Vora YY, Suarez-Almazor M, Steinke DE, Martin ML, Findlay JM. Role of transcranial Doppler monitoring in the diagnosis of cerebral vasospasm after subarachnoid hemorrhage. Neurosurgery 1999; 44: 1237–47 discussion 47–8.

Wadman WJ, Juta AJ, Kamphuis W, Somjen GG. Current source density of sustained potential shifts associated with electrographic seizures and with spreading depression in rat hippocampus. Brain Res 1992; 570: 85–91.

Weidauer S, Vatter H, Beck J, Raabe A, Lanfermann H, Seifert V, et al. Focal laminar cortical infarcts following aneurysmal subarachnoid haemorrhage. Neuroradiology 2008; 50: 1–8.

Woitzik J, Dreier JP, Hecht N, Fiss I, Sandow N, Major S, et al. Delayed cerebral ischemia and spreading depolarization in absence of angiographic vasospasm after subarachnoid hemorrhage. J Cereb Blood Flow Metab 2011. Advance Access published on December 7, 2011, doi:10.1038/jcbfm.2011.169.1.

COSBID-M3: A Platform for Multimodal Monitoring, Data Collection, and Research in Neurocritical Care

J. Adam Wilson, Lori A. Shutter, and Jed A. Hartings

Abstract Neuromonitoring in patients with severe brain trauma and stroke is often limited to intracranial pressure (ICP); advanced neuroscience intensive care units may also monitor brain oxygenation (partial pressure of brain tissue oxygen, $P_{bt}O_2$), electroencephalogram (EEG), cerebral blood flow (CBF), or neurochemistry. For example, cortical spreading depolarizations (CSDs) recorded by electrocorticography (ECoG) are associated with delayed cerebral ischemia after subarachnoid hemorrhage and are an attractive target for novel therapeutic approaches. However, to better understand pathophysiologic relations and realize the potential of multimodal monitoring, a common platform for data collection and integration is needed. We have developed a multimodal system that integrates clinical, research, and imaging data into a single research and development (R&D) platform. Our system is adapted from the widely used BCI2000, a brain-computer interface tool which is written in the C++ language and supports over 20 data acquisition systems. It is optimized for real-time analysis of multimodal data using advanced time and frequency domain analyses and is extensible for research development using a combination of C++, MATLAB, and Python languages. Continuous streams of raw and processed data, including BP (blood pressure), ICP, PtiO2, CBF, ECoG, EEG, and patient video are stored in an open binary data format. Selected events identified in raw (e.g., ICP) or processed (e.g., CSD) measures are displayed graphically, can trigger alarms, or can be sent to researchers or clinicians via text message. For instance, algorithms for automated detection of CSD have been incorporated, and processed ECoG signals are projected onto three-dimensional (3D) brain models based on patient magnetic resonance imaging (MRI) and computed tomographic (CT) scans, allowing real-time correlation of pathoanatomy and cortical function. This platform will provide clinicians and researchers with an advanced tool to investigate pathophysiologic relationships and novel measures of cerebral status, as well as implement treatment algorithms based on such multimodal measures.

Keywords Cortical spreading depolarizations • Electrocorticography • Multimodality monitoring • Neurocritical care • Subarachnoid hemorrhage

Introduction

Delayed cerebral ischemia, including clinical deterioration and the development of new infarcts, is the leading potentially treatable cause of mortality and disability in patients with aneurysmal subarachnoid hemorrhage (SAH). Cerebral vasospasm has been presumed to be a main contributing cause, and periodic assessment of vascular flow by transcranial Doppler and computed tomographic angiography [1] are central to its diagnosis. Methods of continuous monitoring to detect ischemic changes, such as brain tissue oxygenation ($P_{bt}O_2$), cerebral microdialysis [2], quantitative electroencephalography (EEG) [3, 4], thermal diffusion flowmetry [5], and near-infrared spectroscopy [6] have also been investigated. These techniques hold promise not only for early detection of cerebral vasospasm, but also for fresh insight into the pathophysiology of delayed ischemic complications. Results of recent studies have suggested an dissociation between vasospasm and neurologic outcome, prompting a paradigm shift with renewed search for other factors involved in delayed cerebral ischemia [7, 8].

Development and investigation of neuromonitoring methods is therefore critical for understanding cerebral pathophysiology following SAH and for determining best management practices. Unfortunately, however, realization of the full potential of multimodal techniques has been

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limited by the lack of integration of diverse modalities. Each modality typically requires a separate monitor and data collection system; therefore, subsequent downloading, importing, and time synchronizing in an additional system is necessary for multimodal data review. The Component Neuromonitoring System (CNS Technology, LLC, Ambler, PA) is a rare example of a clinical system for integration of multimodal data. However, monitors approved for clinical use are highly restricted in their capabilities to display and process data in new ways, while a system for research requires this ability to prototype and test algorithms for acquisition, processing, display, and real-time decision support. As one example, to discover the clinical meaning and utility of nonseizure EEG activities such as periodic epileptiform discharges, pattern recognition routines could be developed and then displayed alongside processed metrics of other modalities in a common platform. Clearly, advances are limited by our ability to interpret and integrate complex, multimodal data streams.

Our aim was to develop and test a multimodal neuromonitoring system that integrates clinical and research data into a single data collection, analysis, and detection platform. Our specific motivation was to support clinical studies of spreading depolarizations by the CoOperative Study on Brain Injury Depolarizations (http://www.cosbid.org). Spreading depolarizations are pathologic waves of mass neuronal depolarization that are measured by electrocorticography (ECoG) in human cerebral cortex after brain trauma and ischemic and hemorrhagic stroke [9, 10]. In SAH, they are associated with development of delayed cerebral ischemia and infarction and are a potential future target for neuromonitoring and treatment [11–15]. Evidence suggests that spreading depolarizations may be triggered by low blood pressure and high temperature [16], low regional cerebral blood flow (rCBF) [17], and low plasma glucose [18] and may in turn cause changes in cerebral lactate, glucose [18, 19], rCBF [12], and local $P_{bt}O_2$ [20]. To best understand these pathophysiologic relationships, a platform that integrates these multiple modalities is needed. Furthermore, interventional studies using spreading depolarizations as a treatment indication might require a system capable of predicting and detecting their occurrence, monitoring their evolution, and providing user feedback based on depolarizations and possibly criteria from other monitored modalities. A treatment protocol, for instance, might be triggered when depolarizations occur >1/h and CPP is >70 mmHg.

We developed a system, the COSBID-M3, that is adapted from the widely used BCI2000 platform, a brain-computer interface tool for real-time analysis of EEG and ECoG data [21]. BCI2000 has been in use for more than a decade and has been distributed to more than 600 labs worldwide for research. Importantly, it provides a flexible and adaptable research platform capable of performing tasks beyond standard BCI experiments, including functional cortical mapping for epilepsy surgery [22], behavioral experiments in both humans and animals [23], and human-computer interface research. Therefore, we adapted the BCI2000 system to function as a multimodal research monitor capable of acquiring continuous streams of raw data, including arterial blood pressure (ABP), intracranial pressure (ICP), $P_{bt}O_2$, rCBF, EEG, ECoG, and continuous video. It is capable of advanced signal-processing techniques and decision support and is designed in a flexible, open development format. Furthermore, it can be used for researching any signal type or computational algorithm and is not specific to the COSBID group or ECoG signals.
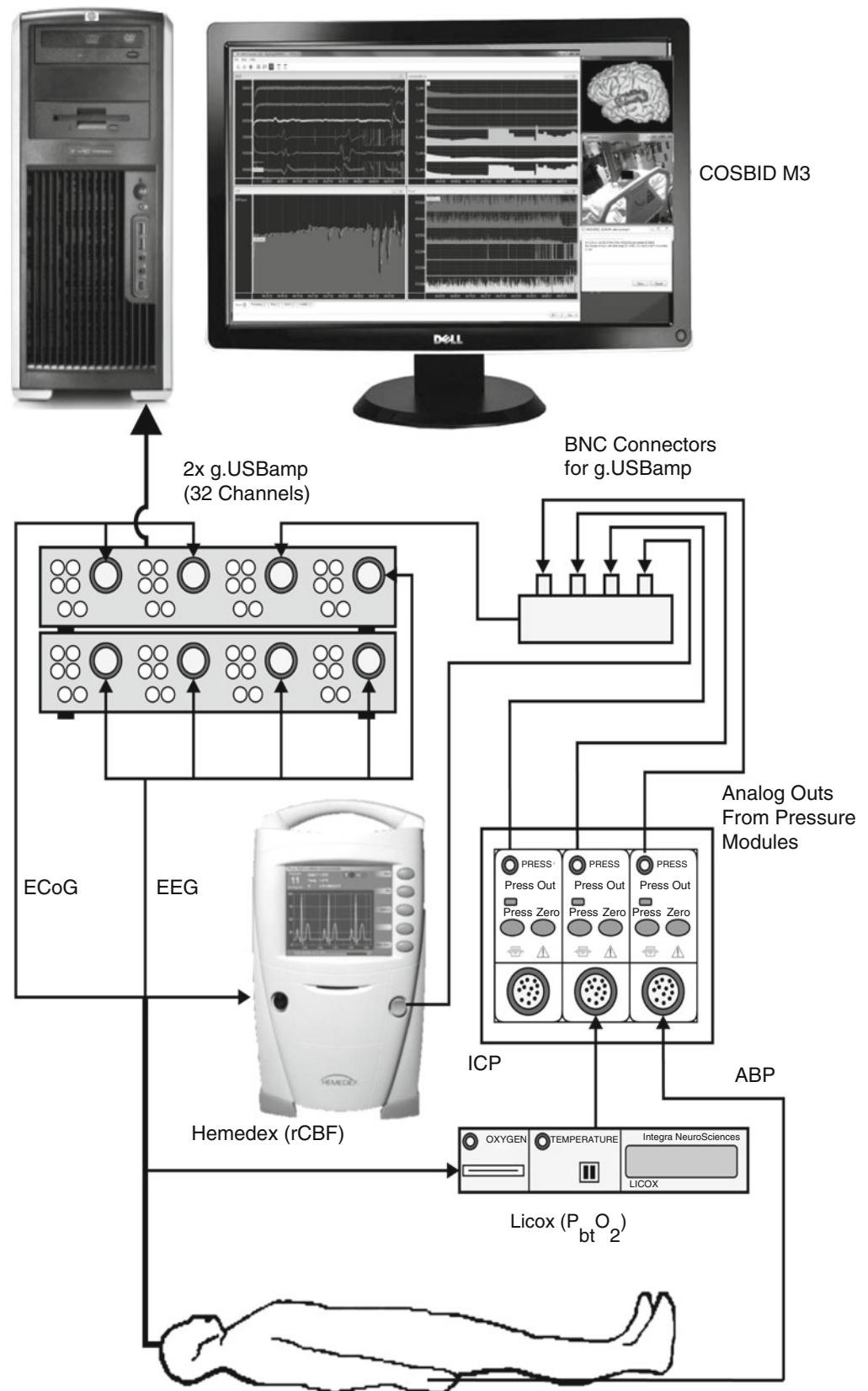
Software System Design

The COSBID-M3 maintains core design principles of the BCI2000 system [21]. These are (1) a common model that can describe any multimodal monitoring system, such that multiple data streams can be split, processed, and recombined to study the interaction of any arbitrary combination of signals; (2) interchangeability and independence of modules, with logical programmatic separation of data acquisition, signal processing, data visualization, system configuration, and decision support; (3) scalability of experimental parameters with no restraints on signal sampling rates, number of channels, or signal-processing complexity; (4) real-time capability such that the time from signal acquisition to processing and display is on the order of milliseconds; and (5) support for offline analysis via a custom data storage format that stores all acquired data, records of all events, and the experimental operating protocol, thus allowing the entire experiment to be replicated offline and in future recording sessions.

The software system is comprised of four independent programs or modules. First, the *Acquisition* module digitizes signals, stores them to disk, and passes them to the processing module. The *Processing* module performs signal analysis routines, ranging from simple mathematical operations (e.g., digital filtering, decimation, matrix multiplication, squaring, etc.) to more complex algorithms (e.g., fast Fourier transforms [FFTs], autoregressive power spectral analysis, seizure detection, etc.). The processed results are passed to the *Decision Support and Feedback* module, which provides useful feedback to the researcher or clinician. The final module is the *Operator*, which handles synchronizing and communication between the other modules, provides an experimental user interface for recording configuration, and handles data visualization (Fig. 1)

Fig. 1 The COSBID M3 system diagram. The M3 acquires signals from various bedside monitors, including arterial blood pressure (*ABP*), intracranial pressure (*ICP*), brain tissue oxygenation ($P_{bt}O_2$), regional cerebral blood flow (*rCBF*), *EEG* (electroencephalogram), and *ECoG* (electrocorticography). EEG and ECoG signals are acquired directly into the g.USBamps. The ABP, ICP, $P_{bt}O_2$, and rCBF are collected from the analog outputs of the pressure sensor modules and Hemedex monitors. These analog signals are input to a box with four BNC connectors. The box uses a voltage divider to decrease the signal amplitude 500 times so that it is in the appropriate input range for the g.USBamp

COSBID M3

2x g.USBamp
(32 Channels)

BNC Connectors
for g.USBamp

ECoG   EEG

Analog Outs
From Pressure
Modules

PRESS   PRESS   PRESS
Press Out   Press Out   Press Out
Press Zero   Press Zero   Press Zero

ICP

ABP

Hemedex (rCBF)

OXYGEN   TEMPERATURE   Integra NeuroSciences
LICOX

Licox ($P_{bt}O_2$)

Data Acquisition

The Acquisition module collects data from one or more sources; currently more than 20 amplifiers and analog-to-digital converters (ADCs) are supported. The primary data source for COSBID studies is full-band ECoG. Therefore, for the core acquisition hardware we have used the g.USBamp (g.tec, Graz, Austria), a 24-bit direct-current amplifier and ADC with a ±250-mV input range and sampling rate of up to 38.6 kHz per channel. Each g.USBamp has 16 analog input channels, and multiple units can be stacked to allow up to 256 channels. Therefore, our basic setup consists of 2 g.USBamps that acquire 6 ECoG channels, 20 EEG channels, ICP, ABP, $P_{bt}O_2$, and rCBF, for a total of 30 data channels (Fig. 1). However, additional channels could be acquired by adding additional g.USBamps or other ADCs.

Data are sampled at configurable rates, which can be set differently for various signals in the same recording. Blocks of data samples are acquired from the ADC, typically every 30–50 ms; for example, a 50-ms block of data sampled at 1,200 Hz will contain 60 samples. The raw, unfiltered data are stored to disk in a single file in the BCI2000 format, sent to the Operator module for visualization, and finally sent to the Processing module. The data file is saved using the BCI2000 format, which is open source and provides interfaces with a number of programming languages, including C++, MATLAB, and Python. The file can also be down-sampled and converted to other formats, such as binary or European data format (EDF), for importing to other data analysis programs such as LabChart (ADInstruments, New South Wales, Australia).

Signal Processing

The Processing module uses a *plug-in* architecture for signal analysis and event detection. Plug-ins serve as algorithmic building blocks that allow complex analysis routines to be constructed. Examples of existing plug-ins are digital filtering, data remontaging, down-sampling, FFT, and threshold detection; new plug-ins may be written using the plug-in framework, described in the following. Prior to monitoring, a signal-processing network of plug-ins is designed in a simple text file in which specific subsets of channels are passed to plug-ins for processing (Fig. 2b). The results of each plug-in are passed to subsequent plug-ins for further processing as needed until all of the designated analyses are complete. For example, to detect depressions of 0.5–100 Hz ECoG brain activity induced by 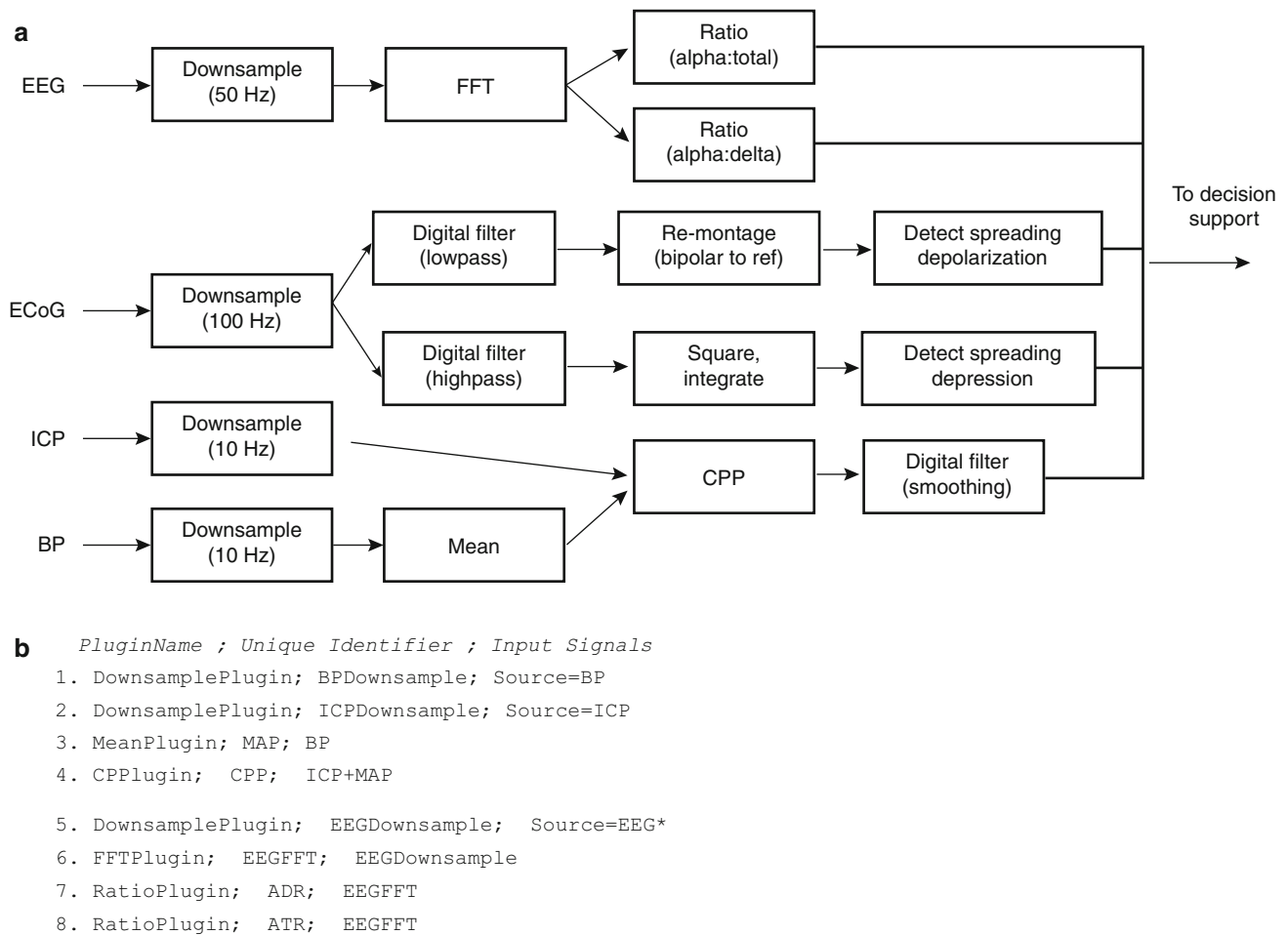spreading depolarizations, a sequence of plug-ins would include a down-sampler, digital filter, squaring and integration filter, and finally a relative threshold filter that detects decreases in this computed power integral. Similarly, cerebral perfusion pressure (CPP) is calculated by building a chain including down-sampling of the ICP and ABP channels, calculating mean values, and finally subtracting ICP from ABP. An example signal-processing chain is shown in Fig. 2a, and a portion of the chain definition is in Fig. 2b.

Although many commonly used plug-ins already exist and are distributed with the COSBID-M3, the plug-in framework was designed to make the M3 extensible and configurable according to specific data-processing needs of individual researchers. Thus, new plug-ins can be created simply using the C++, MATLAB, or Python programming languages. While C++ is the preferred method due to its speed and direct interface with the M3, MATLAB and Python support allows signal-processing prototypes to be tested directly within the real-time system and provides access to the extensive numerical libraries available on these platforms. The M3 framework provides support for data trending, efficient data buffering, and multiple visualization techniques, in addition to many other features. Figure 3 shows an example of the main processing functions for both C++ and MATLAB.

Decision Support and Feedback

The fourth module is the Decision Support and Feedback module. This module is designed to summarize the processed results and relay important information to the researcher or clinicians and caregivers. Using a relatively simple syntax, a variety of displays and alarms can be triggered for any number of conditions based on raw or processed data. For example, to trigger an event when the ICP exceeds 20 mmHg for more than 5 min, the syntax would be ICP >20, 360 s. For a trigger based on multiple conditions, ICP >20 AND CPP <60, 360 s would trigger an event when both ICP is above 20 mmHg and CPP is less than 60 mmHg for longer than 360 s. Another example of using processed data would be detection of decreases in the ECoG power integral, as described previously.

Several default event types have been defined, although these can be extended using the plug-in architecture as well. Currently, events can trigger (1) a visual alert in which flashing text is shown on the display, (2) an audio alert that continuously plays a selected alarm until it is disabled, or (3) an electronic alert that sends an e-mail or text message to a list of recipients. Alerts are useful not

a

b

```
      PluginName ; Unique Identifier ; Input Signals
  1.  DownsamplePlugin; BPDownsample; Source=BP
  2.  DownsamplePlugin; ICPDownsample; Source=ICP
  3.  MeanPlugin; MAP; BP
  4.  CPPlugin;  CPP;  ICP+MAP

  5.  DownsamplePlugin;  EEGDownsample;  Source=EEG*
  6.  FFTPlugin;  EEGFFT;  EEGDownsample
  7.  RatioPlugin;  ADR;  EEGFFT
  8.  RatioPlugin;  ATR;  EEGFFT
```

Fig. 2 (**a**) An example processing chain involving *ECoG* (electrocorticography), *EEG* (electroencephalogram), *ICP* (intracranial pressure), and *BP* (blood pressure). This chain down-samples each data stream appropriately using the *Downsample* plug-in. The EEG is then processed using the *FFT* (fast Fourier transform) plug-in, and the power spectrum from the FFT is passed to the ratio plug-in, which calculates quantitative EEG measures such as the alpha-delta ratio and alpha-total power ratio. The ECoG signal is split into two separate chains that detect both spreading depolarizations and spreading depressions. The ICP and BP are both down-sampled to 10 Hz; the *MAP* (mean arterial pressure http://en.wikipedia.org/wiki/Mean_arterial_pressure) is calculated with a *Mean* plug-in that calculates the mean of the blood pressure over the previous cardiac cycle. The ICP and MAP are recombined in the *CPP* (cerebral perfusion pressure) plug-in, which subtracts the ICP from the MAP to find the CPP. Finally, the CPP is smoothed with a digital filter. It is important to note that many of these plug-ins are reused (e.g., the Downsample and Digital Filter plug-ins), and that the plug-ins do not depend on any particular data type; that is, they are general-purpose plug-ins. (**b**) A portion of the processing definitions for (**a**). *Lines 1–4* define the CPP plug-ins, and *lines 5–8* define the quantitative EEG plug-ins. The general format for a plug-in line is the name of the plug-in, a unique identifier for the plug-in, and a list of input signals, all separated by semicolons. In line 1, the down-sampler is used; it is named "BPDownsample," and it is taking the signal named "BP" as its input. The ICP is down-sampled similarly. On line 3, the output from BPDownsample is passed into the MeanPlugin, which calculates the mean over some period and is named MAP. Finally, on line 4 the CPPPlugin, named CPP, takes the ICP and MAP signals as input to find the CPP. On line 5, the DownsamplePlugin, named EEGDownsample, takes all channels with "EEG" in the name as input; the "*" character is used as a wild card for string matching, so that channels named EEG1, EEG2, … , EEG20 will all match as input for this plug-in. On line 6, the FFTPlugin takes the FFT of all EEG channels over a specified duration (e.g., 10 s). On lines 7 and 8, the QEEG plug-ins are defined for the alpha-delta ratio (*line 7*) and alpha-total power ratio (*line 8*)

only for routine clinical care but also particularly for research protocols. They could be used, for instance, to alert staff to collect more detailed clinical data surrounding particular events in observational research or to activate a treatment protocol in an interventional trial that targets neuromonitoring variables. As a practical benefit, they can also alert staff to malfunctions or unintended data interruptions.

a
```
    void
    CPPplugin::Process (const GenericSignal& Input,
                        GenericSignal& Output)
    {
1....  float mCPP = Input (mMAPch,0) - Input (mICPch,0);
2....  mCPPtrend. Insert (mCPP);
3....  Output (0,0) = mCPP;
4....  mCPPvis . Send (Output);  // send results to graph
    }
```

b
```
function out_signal = bci_Process (in_signal)
out_signal = in_signal * refToBipolarMatrix;
```

Fig. 3 Example plug-in code written in C++ (**a**) and MATLAB (**b**). (**a**) The *Input* signal contains the mean arterial pressure (MAP; calculated in the preceding plug-in) and the intracranial pressure (ICP). This plug-in calculates the cerebral perfusion pressure by subtracting the ICP channel from the MAP channel (*line 1*). The CPP (cerebral perfusion pressure) is inserted into a trending buffer (*line 2*) and written to the Output signal (*line 3*). Last, the *Output* signal is sent to the visualization window for the CPP. (**b**) This MATLAB function rereferences the input signal (*in_signal*) using matrix multiplication. The result is saved to *out_signal* in a single line of code and returns the result to the M3

Configuration and Data Visualization

The Operator module handles the most common user interface and visualization tasks. Prior to starting recording, the system is set up via a Configuration Tool, in which hundreds of variables describing the experiment are set. These variables typically have intelligent preset values, and the default values are loaded from a configuration file. Therefore, once a standard experimental and monitoring paradigm is defined and saved in the configuration file, the setup time can be rapid, requiring only entry of subject-specific information (e.g., subject identifiers). In addition, a graphical tool is provided to configure new signals, processing chains, and decision support quickly.

The Acquisition and Processing modules send processed signals to the Operator for visualization. This can consist of raw time series data or any processed data as computed by the Processing module. The monitoring display contains all of the visualization and log windows (Fig. 4). These windows are grouped into tabs for organization and can be moved from tab to tab to compare multiple data streams. For example, Fig. 4 shows three different processed ECoG windows (low-pass filtered, high-pass filtered and squared, and the alpha-delta ratio for each channel) along with the CPP. There are many options for configuring the displays, such as different colors, line fills, and signal clipping, and axes can be changed to display the desired amplitudes and timescales. Elementary data processing, such as filtering and squaring, is also available; this processing is performed for visualization only, does not interrupt data acquisition or affect recorded data, and does not need to be set in the Configuration tool.

Conclusion

The COSBID-M3 provides researchers with a flexible, real-time system for multimodal data acquisition, processing, and display at the bedside. Over 20 amplifier systems are currently supported, and any system that provides a programming interface can be added as well. The g.USBamp was chosen for its previously discussed acquisition characteristics designed for EEG and ECoG; however, alternative, less-expensive amplifiers such as those available from DataTranslation could be used instead for other research purposes, such as computation of autoregulatory indices, rapid microdialysis [24], near-infrared spectroscopy, or laser Doppler flowmetry.

Development of the M3 is ongoing, and future versions will include a full review system integrated with the signal-processing frame to allow review of processed data, inclusion of additional signals such as microdialysis, and better incorporation of imaging results. We also plan to add support for efficient annotations at the bedside, including clinical exam notes, comments, and other notes and results. The COSBID-M3 system is freely available for download. More information may be found at http://www.cosbidm3.com. We are actively seeking developers to contribute to this project and need clinicians to help drive development. Therefore, this highly customizable and extensible platform will provide clinicians and researchers with an advanced tool to investigate pathophysiologic relationships and implement advanced multimodal treatment algorithms.
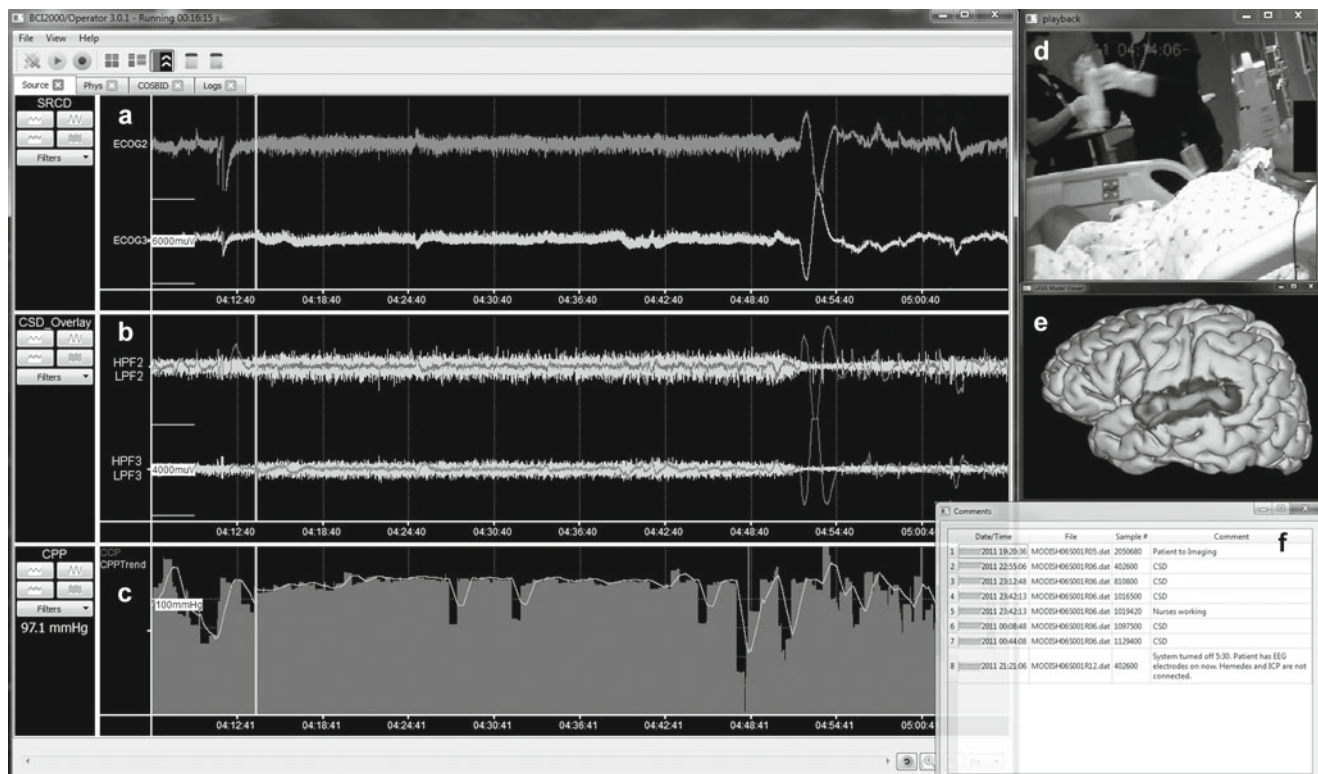
Fig. 4 Example display during monitoring. The main display shows two raw ECoG signals during a spreading depolarization (**a**), the same two channels with the low-pass filtered signal overlaid on the high-pass filtered signal (**b**), and the cerebral perfusion pressure (**c**). The secondary windows on the right contain the synchronized patient video (**d**), the 3D MRI model co-registered with CT data (**e**), and the comment and event log (**f**)

References

1. Washington CW, Zipfel GJ (2011) Detection and monitoring of vasospasm and delayed cerebral ischemia: a review and assessment of the literature. Neurocrit Care 15(2):312–317
2. Macdonald RL (2009) Subarachnoid hemorrhage and microdialysis. J Neurosurg 111(5):907–908
3. Vespa PM et al (1997) Early detection of vasospasm after acute subarachnoid hemorrhage using continuous EEG ICU monitoring. Electroencephalogr Clin Neurophysiol 103(6):607–615
4. Labar DR et al (1991) Quantitative EEG monitoring for patients with subarachnoid hemorrhage. Electroencephalogr Clin Neurophysiol 78(5):325–332
5. Vajkoczy P et al (2003) Regional cerebral blood flow monitoring in the diagnosis of delayed ischemia following aneurysmal subarachnoid hemorrhage. J Neurosurg 98(6):1227–1234
6. Zweifel C et al (2010) Continuous assessment of cerebral autoregulation with near-infrared spectroscopy in adults after subarachnoid hemorrhage. Stroke 41(9):1963–1968
7. Pluta RM et al (2009) Cerebral vasospasm following subarachnoid hemorrhage: time for a new world of thought. Neurol Res 31(2):151–158
8. Hansen-Schwartz J et al (2007) Cerebral vasospasm: looking beyond vasoconstriction. Trends Pharmacol Sci 28(6):252–256
9. Lauritzen M et al (2011) Clinical relevance of cortical spreading depression in neurological disorders: migraine, malignant stroke, subarachnoid and intracranial hemorrhage, and traumatic brain injury. J Cereb Blood Flow Metab 31(1):17–35
10. Dreier JP (2011) The role of spreading depression, spreading depolarization and spreading ischemia in neurological disease. Nat Med 17(4):439–447
11. Dreier JP et al (2006) Delayed ischaemic neurological deficits after subarachnoid haemorrhage are associated with clusters of spreading depolarizations. Brain 129(Pt 12):3224–3237
12. Dreier JP et al (2009) Cortical spreading ischaemia is a novel process involved in ischaemic damage in patients with aneurysmal subarachnoid haemorrhage. Brain 132(Pt 7):1866–1881
13. Iadecola C (2009) Bleeding in the brain: killer waves of depolarization in subarachnoid bleed. Nat Med 15(10):1131–1132
14. Hartings JA et al (2011) Spreading depolarizations have prolonged direct current shifts and are associated with poor outcome in brain trauma. Brain 134:1529–1540
15. Dohmen C et al (2008) Spreading depolarizations occur in human ischemic stroke with high incidence. Ann Neurol 63(6):720–728
16. Hartings JA et al (2009) Spreading depolarizations and late secondary insults after traumatic brain injury. J Neurotrauma 26(11): 1857–1866

17. Nakamura H et al (2010) Spreading depolarizations cycle around and enlarge focal ischaemic brain lesions. Brain 133(Pt 7):1994–2006

18. Hopwood SE et al (2005) Transient changes in cortical glucose and lactate levels associated with peri-infarct depolarisations, studied with rapid-sampling microdialysis. J Cereb Blood Flow Metab 25(3):391–401

19. Krajewski KL et al (2011) Cerebral microdialysis in acutely brain-injured patients with spreading depolarizations. Acta Neurochir Suppl 110(Pt 1):125–130

20. Bosche B et al (2010) Recurrent spreading depolarizations after subarachnoid hemorrhage decreases oxygen availability in human cerebral cortex. Ann Neurol 67(5):607–617

21. Schalk G et al (2004) BCI2000: a general-purpose brain-computer interface (BCI) system. IEEE Trans Biomed Eng 51(6):1034–1043

22. Brunner P et al (2009) A practical procedure for real-time functional mapping of eloquent cortex using electrocorticographic signals in humans. Epilepsy Behav 15(3):278–286

23. Felton EA et al (2009) Evaluation of a modified Fitts law brain-computer interface target acquisition task in Able and motor disabled individuals. J Neural Eng 6(5):056002

24. Feuerstein D et al (2010) Dynamic metabolic response to multiple spreading depolarizations in patients with acute brain injury: an online microdialysis study. J Cereb Blood Flow Metab 30(7):1343

A matter of perspective: detection of spreading depolarizations by continuous EEG

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Introduction

Cortical spreading depolarizations are a primary mechanism of secondary injury after stroke and severe traumatic brain injury (TBI) and are associated with poor clinical outcomes.[1] Conventional wisdom for over 60 years was that depolarization waves can not be detected by scalp electroencephalography (EEG), but recent evidence suggests otherwise.[2]

Methods

Thirteen patients with severe TBI were enrolled in a study of spreading depolarizations, measured by electrocorticography (ECoG) from electrode strips placed on the surface of the brain adjacent to the injury focus. Only patients with a clinical need for surgery were enrolled. After surgery, ECoG was recorded during intensive care, as previously described,[1] and scalp EEG was recorded as clinical standard-of-care following the local protocol for multi-modal monitoring of severe TBI. Sixteen scalp electrodes were placed bilaterally according to the international 10-20 standard and were acquired in a 'double-banana' bipolar montage with a Grass amplifier system (0.5 Hz high-pass). Depolarizations were first identified on ECoG by slow potential changes (0.01-0.1 Hz) and depressions of 0.5-50 Hz activity propagating between electrodes.[3] ECoG/EEG signals were then merged for EEG analysis. Leaky power integrals (120 s decay time constant) were calculated for quantification of EEG changes. Research protocols were IRB-approved.

Results

ECoG and EEG were recorded for an average 5.0 and 3.4 days, respectively, and the total duration of simultaneous recordings was 39.5 days. A total of 427 spreading depolarizations occurred in 11/13 patients on ECoG, but only 164 occurred during simultaneous ECoG/EEG in 8 patients. All 8 patients had moderate-to-severe generalized slowing on EEG and 6 had continuous polymorphic delta activity localized to the injured hemisphere.

EEG correlates of ECoG depolarizations were assessed by visual inspection on a highly compressed time scale of 40-90 mm per hour. EEG amplitude depressions occurred in association with 61 (37%) of 164 ECoG depolarizations in up to 3 bipolar EEG channels closest to the ECoG electrode strip. The maximal depression of EEG power relative to baseline was 55% (median; inter-quartile range: 44-63), which developed progressively over a period of 12 min (IQR: 8-17). In most cases (53/60), EEG amplitude recovered prior to subsequent ECoG depolarizations. For these, the total time envelope from start of depression to recovery to a steady-state amplitude was 17 min (IQR:13-24).

Of the remaining 103 ECoG depolarizations that did not induce EEG depression periods, 58 occurred during a period of continued maximal depression (n=15) or partial depression with incomplete recovery (n=43) after a prior depolarization. Thus, these instances exhibited a fusion of EEG depression periods induced by multiple repetitive depolarizations; the intervals between these depolarizations were only 36 min (IQR: 24-47). This is significantly shorter than intervals preceding depolarizations that were associated with EEG depressions (88 min; IQR: 47-111, $p<0.001$). Nineteen ECoG depolarizations without EEG depression could not be explained by persisting depression periods, but also occurred at short intervals (39 min; IQR: 31-46). Finally, 26 depolarizations occurred at the start of EEG recordings so that baseline amplitudes could not be determined.

Conclusions

We found that 37% of spreading depolarizations detected by the gold standard of ECoG are evidenced on EEG by inducing amplitude depressions to 55% of baseline power. An additional 35% of spreading depolarizations were evidenced in continuing depression periods that, after these initial amplitude reductions, persisted through a series of depolarizations occurring at short intervals. Initial EEG amplitude depressions developed over a period of 5-30 min, in contrast to <1 min in ECoG. This is likely due to the broad spatial sampling (~10 cm^2) of EEG electrodes and the time required for a depolarization spreading through cortex at 1-5 mm/min to traverse and depress a sufficient portion of this area. Thus, detection of spreading depolarizations by EEG has been possible since their discovery in 1944[4] and is simply a matter of perspective: EEG must be viewed on a highly compressed time scale to appreciate these slow, creeping changes.

References

1.   Hartings JA, Bullock MR, Okonkwo DO, Murray LS, Murray GD, Fabricius M, et al. Spreading depolarisations and outcome after traumatic brain injury: a prospective observational study. Lancet Neurol. 2011; **10**(12): 1058-64.
2.   Drenckhahn C, Winkler MK, Major S, Scheel M, Kang EJ, Pinczolits A, et al. Correlates of spreading depolarization in human scalp electroencephalography. Brain. 2012; **135**(Pt 3): 853-68.
3.   Fabricius M, Fuhr S, Bhatia R, Boutelle M, Hashemi P, Strong AJ, et al. Cortical spreading depression and peri-infarct depolarization in acutely injured human cerebral cortex. Brain. 2006; **129**(Pt 3): 778-90.
4.   Leao AAP. Spreading depression of activity in the cerebral cortex. J Neurophysiol. 1944; **7**: 359-90.

Pre-emptive decompressive craniectomy: a comparative effectiveness study between a UK and US center

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Introduction

The uses of decompressive craniectomy as either a primary pre-emptive procedure or as a secondary procedure indicated by refractory elevated intracranial pressure (ICP) are controversial in treatment of traumatic brain injury (TBI). A majority of candidates for surgery fall between these extremes and comparative effectiveness studies may elucidate best practices for such real-world scenarios.

Methods

We compared outcomes and surgical methods to treat TBI between two neurosurgical centers in the U.S. and U.K. Only patients with a clinical need for surgery were enrolled, and those with fixed, dilated pupils were excluded. Research protocols were approved by ethical boards. To measure injury severity, we used the prognostic score based on seven variables collected prospectively at hospital admission, as defined by the IMPACT study.[1, 2] Electrocorticographic recordings were collected during intensive care using a subdural electrode strip placed during surgery, as described previously,[3] and ICP was measured when clinically indicated. Operative notes were reviewed to determine the primary indications for surgery and the surgical procedures performed. Post-surgical CT scans were evaluated by a neurosurgeon blinded to other data for quantification of craniotomy/-ectomy type and size. Bone flap areas were approximated using the formula for an ellipse. Six-month neurologic outcome was assessed by the Glasgow Outcome Scale (GOS).

Results

Patients with surgical treatment of TBI were enrolled in the Co-Operative Study on Brain Injury Depolarizations at King's College Hospital (KCH, London, UK; n=27) and Virginia Commonwealth University (VCU, Richmond, VA; n=24) from 2004-2010. Baseline measures of severity were similar for VCU and KCH, including prognostic scores based on admission characteristics (p=0.22), maximal pre-operative ICP values (48 vs 44 mmHg, respectively, p=0.67), and indications for surgery (initial CT as primary factor in 58% and 56% of cases, respectively). However, at VCU patients were operated earlier (83% vs 52% within 24 h, p=0.02), craniectomies with removal of bone flap were performed more frequently (75% vs 44%, p=0.03) and craniotomy/-ectomy areas were 56% larger (mean: 82 vs 53 cm^2, p<0.001). Post-operatively, VCU patients had lower maximal ICP values (22.5 vs 31.4 mmHg, p<0.01) and trended toward lower incidence of cortical spreading depolarizations (42% vs. 63%, p=0.13) and more favorable outcomes (GOS 3-5; 67% vs. 46%, p=0.14).

In order to determine whether differences are attributable to the use of prophylactic decompressive craniectomy at VCU, as opposed to smaller craniotomies for lesion evacuation only at KCH, we also compared only those patients requiring lesion evacuation within 24 h after injury. For 16 VCU and 14 KCH patients, prognostic scores, pre-operative ICP values, and timings of surgeries were similar, and the majority of both groups had subdural hematomas

evacuated (VCU: 14/16 and KCH: 12/14). However, differences in surgical procedures and post-operative courses were even more accentuated in this subgroup. At VCU, craniectomies were performed more frequently (75% vs 36%, p=0.03) and craniotomy/-ectomy areas were 78% larger (mean: 85 vs 48 cm^2, p<0.001). Following surgery, VCU patients had lower maximal ICP values (20.8 vs 30.2 mmHg, p=0.02) and lower incidences of cortical spreading depolarizations (31% vs. 86%, p<0.01). Accordingly, VCU patients had significantly more favorable outcomes (69% vs 29%, p=0.03).

Conclusions

Our results confirm signficant variability in surgical management of TBI, with surgeries performed earlier, craniotomies larger, and bone flaps removed more frequently at a U.S. compared to a European center. These differences were even more accentuated in patients undergoing emergency surgery for lesion evacuation, and were associated with less post-operative intracranial pathology, evidenced by raised ICP and spreading depolarizations, and better outcomes at the U.S. center. Results support the use of early prophylactic decompressive craniectomy as a primary surgical intervention in patients with mass lesions. Furthermore, they illustrate the use of spreading depolarizations as a biomarker of intracranial pathology and indicator of worse outcome.[3, 4] A limitation of the study is that undocumented differences in clinical care may have contributed to the different outcomes observed. Nevertheless, such observational and comparative effectiveness research has advantages over controlled clinical trials in identifying best real-world management practices and may assist guideline development.

References

1. Murray GD, Butcher I, McHugh GS, Lu J, Mushkudiani NA, Maas AI, et al. Multivariable prognostic analysis in traumatic brain injury: results from the IMPACT study. J Neurotrauma. 2007; **24**(2): 329-37.
2. Hukkelhoven CW, Steyerberg EW, Habbema JD, Farace E, Marmarou A, Murray GD, et al. Predicting outcome after traumatic brain injury: development and validation of a prognostic score based on admission characteristics. J Neurotrauma. 2005; **22**(10): 1025-39.
3. Hartings JA, Bullock MR, Okonkwo DO, Murray LS, Murray GD, Fabricius M, et al. Spreading depolarisations and outcome after traumatic brain injury: a prospective observational study. Lancet Neurol. 2011; **10**(12): 1058-64.
4. Dreier JP, Woitzik J, Fabricius M, Bhatia R, Major S, Drenckhahn C, et al. Delayed ischaemic neurological deficits after subarachnoid haemorrhage are associated with clusters of spreading depolarizations. Brain. 2006; **129**(Pt 12): 3224-37.